UNIVERSIDAD AUTÓNOMA DE MADRID

TESIS DOCTORAL

Analysis of Social Behavior in Zebrafish

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Table of contents

List of figures

Summary

In this thesis, I have studied a number of aspects of social and collective behavior in zebrafish. Zebrafish (*Danio Rerio*) is a model organism used in a variety of different fields of research. While larval zebrafish are used in neurobiology and genetics, adult animals serve as a model for social and collective behavior.

In order to combine these two applications and use zebrafish for the study of the neurological basis of social behavior in vertebrates, the first objective of this thesis was to determine if there is a period of time in the development of zebrafish during which animals have already developed sufficiently to show clear social interactions, while at the same time they have not lost the larval characteristics which allow opto-genetic and neurobiological manipulation and imaging techniques.

The second objective was the development and study of methods for the characterization of behavioral changes which occur during development in groups of zebrafish.

Finally, I propose a mathematical model which describes the emergence of collective behavior in zebrafish larva on the basis of social interactions between each individual and the group.

In Chapter 1, I present *idSocial*, a Matlab® toolbox for the analysis of individual and social behavior. Zebrafish shows a great behavioral repertoire ranging from shoaling and schooling behavior over aggression and dominance to territorial behavior. *idSocial* includes methods for the analysis of a number of different aspects of these behaviors. Examples of the variety and the utility of available methods are given using real data from free-swimming fish:
The behavior of a zebrafish strain is compared to a control group using basic measures like individual speed and acceleration. The difference between shoaling and schooling behavior is shown presenting results from zebrafish and Medaka fish. Leader-Follower hierarchies in schooling behavior are presented for Medaka. The attraction of an animal towards a conspecific depending on its relative position is given as an example of how to determine individual rules of interaction of individuals in a group. The emergence of territories, connected with

fights and chases between individuals, is presented for a group of five zebrafish.
Finally, an extensive example of the practical use of *idSocial* is given in Chapter 2.

In Chapter 2, I study the development of social behavior in zebrafish larvae during the first three weeks of their life. I used an experimental setup which minimizes external, non-social stimuli and provides the conditions necessary for the automated extraction of coordinates using the software *idTracker*. By applying a series of methods, I have shown that on the population level robust social behavior is present in zebrafish larvae from 8 days post-fertilization on, and can be detected even earlier for single individuals. At this age, neurogenetic imaging and manipulation techniques can still be applied in order to study the neurological processes involved in social behavior in vertebrates.

The results show that social behavior in Zebrafish increases gradually over the course of the first weeks of development. This increase in social behavior is characterized by a decrease in inter-individual distance, an increasing probability of an individual turning and accelerating towards a conspecific, and increasingly long periods of time which animals spend close to each other without leaving the group. The techniques that I used do not only allow the measurement of social behavior for the group as a whole but for each individual in the group separately, and already at an age of 6 days post fertilization I find individuals which show significant attraction towards their conspecifics.

Resumen

En esta tesis, he estudiado una serie de aspectos de comportamiento social y colectivo en pez cebra. El pez cebra (*Danio Rerio*) es un organismo modelo que se usa en una variedad de campos de investigación. Mientras que el estado de larva se utiliza para estudios de neurociencia y genética, el adulto se usa como modelo para el comportamiento social y colectivo.

Para poder combinar los dos aspectos y usar pez cebra para entender el fundamento neurológico del comportamiento social en vertebrados, el primer objetivo de esta tesis consiste en determinar si existe un periodo de tiempo en el desarrollo de pez cebra en el que los animales ya están lo suficientemente desarrollados para mostrar interacciones sociales, mientras que no hayan perdido las características presentes en larvas que permiten el uso de técnicas opto-genéticas y neurobiológicos.

El segundo objetivo es el desarrollo y el estudio de métodos para la caracterización de cambios en el comportamiento que ocurren durante el desarrollo en grupos de pez cebra.

Por último, propongo un modelo matemático que describe la aparición del comportamiento colectivo en larvas de pez cebra basándose en las interacciones entre cada individuo y el grupo.

En el capítulo 1, presento *idSocial*, una colección de herramientas de Matlab® para el análisis de comportamiento individual y social. El pez cebra muestra un gran repertorio de comportamientos, incluyendo la agrupación social ('shoaling'), el movimiento coordinado ('schooling'), agresión y comportamiento dominante y territorial. *idSocial* incluye métodos para el análisis de una serie de aspectos diferentes de estos comportamientos. Presento ejemplos de la variedad y utilidad de los métodos disponibles usando datos reales de experimentos con peces que nadaban libremente:
Comparo el comportamiento de una cepa de pez cebra con un grupo de controles usando medidas básicas como la velocidad y aceleración. Muestro la diferencia entre el movimiento libre ('shoaling') y el movimiento coordinado ('schooling') del grupo mediante resultados

obtenidos para pez cebra y Medaka (*Oryzias latipes*). Presento un análisis de las relaciones líder-seguidor en el movimiento coordinado de Medaka. Como ejemplo para las reglas de interacción de individuos en un grupo, presento la atracción de un animal hacía un congénere en función de su posición relativa. Estudio la aparición de territorios en conexión con luchas y persecuciones entre individuos de un grupo de 5 animales. Por último, doy un ejemplo extenso del uso práctico de *idSocial* en el capítulo 2.

En el capítulo 2, estudio el desarrollo del comportamiento social en larvas de pez cebra durante las primeras tres semanas de vida. Usé un montaje experimental que minimizaba los estímulos externos y no sociales y ofrecía las condiciones necesarias para la extracción automática de las coordenadas usando el software *idTracker*. Aplicando una serie de métodos, he mostrado que a nivel de la población se aprecia comportamiento social robusto en larvas de pez cebra a partir de una edad de 8 días post-fertilización, y que se puede detectar incluso antes para algunos individuos. A esta edad, aún se pueden aplicar técnicas neuro-genéticas para la visualización y manipulación con el fin de estudiar los procesos neurológicos involucrados en el comportamiento social en vertebrados.

Los resultados muestran que el comportamiento social en pez cebra aumenta gradualmente a lo largo de las primeras semanas del desarrollo. Ese aumento se caracteriza por una disminución de las distancias entre los individuos, un aumento de la probabilidad de que un individuo gire y acelere hacia un congénere, y periodos crecientes del tiempo que los individuos pasan cerca sin salir del grupo.

Las técnicas que he usado no solo permiten medir el comportamiento social de un grupo entero, sino también de cada individuo por separado, y tan temprano como el día 6 post-fertilización se encuentran individuos que muestran una atracción social significativa más fuerte que la que se esperaría a base de los datos control.

General Introduction

Social behavior can be observed throughout the animal kingdom, manifesting itself in a variety of forms and over a broad range of species. The specific practical advantages species gain by living in groups determine the social and collective phenomena we see in nature: Animals look for the proximity of their conspecifics in order to protect themselves from predators, share information about food sources or form packs in order to maximize hunting success. In the long course of evolution, thus emerged the coordinated movement in flocks of birds and shoals of fish, or the cooperative hunting techniques of wolves, strategies that proved successful for the survival of the respective species.

More sophisticated social behavior can be found in eusocial societies, characterized by the cooperative care of the offspring and division of labor among individuals as found in some insect species like ants and bees, and, ultimately, humans.

Due to this ubiquitous presence of social phenomena, their complexity and beauty, and, finally, their meaning for human life, the study of social behavior has ever since attracted interest of a variety of scientific fields, ranging from ecology and ethology over biomedical research and pharmacology, molecular and developmental genetics to statistical physics. The generality of socially motivated organization of animal groups on the one hand motivates the search for universal principles of social interaction and the mechanisms of self-organization in collective behavior. On the other hand it offers the possibility of taking animals as model organisms for the study of social behavior and its underlying physiological aspects.

We want to understand how social behavior arises and what shapes it can take. How do social interactions with others influence our own behavior? And how do interactions between individuals influence the behavior of a collective? Even though these two questions are closely related to each other, they can be looked at from two different perspectives: The first refers predominantly to individual behavior in a social context. The second on the other hand deals with the emergence of collective behavior on the basis of the constituents of a group. In order to fully understand this complex interplay between the individual and the group,

a number of aspects has to be taken into account: What rules of interaction do individuals of a group follow? What kind of social information do individuals use, and how do they use it? How do individual decisions influence in the decision of the group? To answer these questions, we need to study the physiological basis of social behavior in the individual, the role of the nervous system in perception, information integration and decision making. And we need to study the rules by which collective phenomena emerge from the interaction between individuals.

Thus, the prerequisites for a methodical, comprehensive study of social behavior in all its aspects are an adequate, social model organisms, appropriate genetic tools, experimental paradigms, tools for data acquisition and methods of analysis. Especially the recent advances in genetics and neurobiology have opened up the possibility of studying the neural and genetic basis of social behavior. Many of the recent techniques for brain imaging and manipulation have been developed for a small vertebrate model species, zebrafish (*Danio Rerio*). Zebrafish, a small tropical freshwater fish originally found in the rice fields and floodplains of India and Bangladesh (Engeszer et al. [14]), is now widely used as a model organism in molecular genetics, developmental biology and medical research. Due to its rich repertoire of behavioral traits it has also emerged as a model in behavioral research in recent years, making it ideal for multidisciplinary research.

Zebrafish show a wide range of highly adaptive behaviors, with which they react to changes in their environment. Each scientific field tends to ask their own specific questions about zebrafish behavior. Ecology and evolutionary biology look at behavioral traits in order to put them in context with the environment they have developed in. In developmental biology, the aim of measuring zebrafish behavior is to find the effect of mutations or drugs on certain behavioral traits, ranging from basic motor behaviors over preference tasks to the study of social interaction. Noeuroethology deals with the role of the nervous system in animal behavior.

The rather recent field of animal collective behavior has started to use zebrafish as a model to study the rules of interaction, the mechanisms by which groups form, coordinate and take collective decisions. Here, the experience and tools developed for zebrafish can be used in the opposite way: Instead of manipulating zebrafish behavior in order to find the physiological origins of social behavior, the focus lies on how the manipulation of individual behavior changes the dynamics of a group.

However, while there exists a great variety of sophisticated tools for genetic and neurobiological manipulation and cell- and neuroimaging in zebrafish, tools for the quantification and analysis of behavioral data are still rare, especially when it comes to the study of social behavior. The combined study of social interactions in zebrafish and their genetic and neural basis involves several difficulties:

Genetic and neurobiological manipulation and imaging is exclusively done in zebrafish larvae, mainly because they are transparent at a young age, but lose this characteristic during development. However, there are few experimental paradigms for the measurement of social behavior in young zebrafish, especially when it comes to the study of free-swimming, freely interacting individuals. This is mainly due to the technical difficulties involved in measuring the positions and movements of individual larvae.

With *idTracker* (Pérez-Escudero et al. [30]) only recently has emerged a tool which is able to allows for the reliable extraction of positional data of each individual and throughout an experiment, and thus lift some of the difficulties.

In this thesis, we show that *idTracker* is able to track free-swimming zebrafish larvae. We present a toolbox for the analysis of social behavior, *idSocial*, and show how it can be used to analyze naturalistic behavior of free-swimming fish, exploiting the possibilities offered by *idTracker*. We carefully asses changes of social behavior in groups of zebrafish larvae, and we propose a mathematical model which allows a possible interpretation of the observed mechanisms of aggregation in free-swimming zebrafish.

Chapter 1

Analyzing Behavior of Free-Swimming Fish using *idSocial*

1.1 Introduction

The rise of modern genetics and neurobiology brought about the need for small, easily reproducible model species which permits a large number of experiments and data acquisition. In the multidisciplinary field of neuroethology, the same animal species are used to study animal behavior and its underlying mechanisms in the nervous system. However, since in general both the underlying physiology and the connected behavior involve a high degree of variability, tools are needed which facilitate the acquisition and analysis of a big volume of data.

Advances in computer vision made over the last years help to automate and accelerate the quantification and analysis of animal behavior in the laboratory. Especially useful tools have been developed to extract the trajectories of moving animals from previously recorded videos (e.g., Branson et al. [6],Miller and Gerlai [23]). These tools already broaden the range of possible experiments by allowing greater numbers of experiments of higher duration, but often need manual correction which makes them unsuitable for bigger data sets. A recently developed tracking tool, *idTracker* (Pérez-Escudero et al. [30]), is capable of automatically tracking and maintaining the identities of each individual of a group throughout the course of an experiment, making it possible to ask additional questions about individual behavior and its influence on the behavior of the collective.

The possibilities offered by these recent developments have yet to be fully exploited. For example, instead of studying the effect of a specific drug or genetic modification on individual behavior, it is now possible to go the opposite direction and to investigate the influence of a specific individual behavior on the behavior of a whole group, and in this way to deduce conditions and characteristics of social and collective behavior.

Traditionally, social behavior is analyzed in terms of simple quantities like inter-individual the distance between groups members or the polarization of a group (Clark and Evans [10], Buske and Gerlai [8],Miller and Gerlai [24]), but more elaborate measurements are emerging due to the new quality of experimental data obtained by automated tracking systems. Here, we introduce *idSocial*, a Matlab® toolbox providing easy-to-use functions for the analysis of individual and social behavior. It was designed as an extension of *idTracker*, but can also work with trajectories obtained by other tools.*idSocial* is an easily expandable platform which allows the analysis of big data sets.

Zebrafish have a rich repertoire of different behaviors with which they react to changes in their environment. While showing social and cooperative behavior when looking for food sources, exploring an unknown environment or facing predators, they also form dominance hierarchies involving agonistic and aggressive behavior when defending a food source, form territories or fight for a mating partner.

All of these behaviors can be observed in arenas which allow the animals to swim freely and to interact naturally with their conspecifics. By using *idTracker*, the full trajectory for each individual can be obtained. These trajectories contain a great amount of information about social and non-social behavior. This chapter gives an overview over a variety of behaviors in zebrafish and their analysis with *idSocial*.

In the following sections, we will describe and demonstrate a variety of methods to analyze fish behavior. All analyses have been carried out with *idSocial*. The main model organism will be, as throughout this thesis, zebrafish *Danio Rerio*. The Japanese rice fish (*Oryzias latipes*), also known as medaka, will serve as a comparative model to highlight the diversity and differences between species.

Even though our main focus lies on social and collective behavior, we will start by presenting methods to analyze individual behavior of both isolated animals and individuals which are members of a group. The analysis of individual behavior of zebrafish is used for example in order to study the effects of drugs or genetic modifications on locomotion (Pietri et al. [32],Burgess and Granato [7]), but is also of importance when concerned with

collective phenomena. Firstly, individual behavior in terms of the swimming ability and individual preferences of a single animal form the first building block of group movement. And secondly, in order to come to a closer understanding of collective behavior and emergent properties in animal groups, it is fundamental to study the differences of an individuals' behavior when it is isolated or member of a group.

Following the analysis of individual behavior We give an overview of the most prominent social behaviors in zebra fish and their analyses using *idSocial*. We start with those types of social behavior which are characterized by a high degree of aggregation, comparing dynamical structures with a high degree of organization ("schooling"), with less synchronized structures ("shoaling") (see Section 1.3). Eventually, we will give an example of the transition from social to aggressive and territorial behavior (Section 1.5).

1.2 Individual Behavior

In the following, individual behavior refers to the behavior of an individual animal, as opposed to a behavior which is measured taking into account the presence of other conspecifics. This does not mean that the data which is to be analyzed has to come from an isolated animal, but that the quantities we are looking at do not require the presence of other animals. Moreover, we deal with quantities which can be deduced directly from the trajectory of the animal, like the speed of travel, acceleration or deceleration, while not measuring directly the underlying motor behaviors and body dynamics.

Both social and individual behavior can be very variable as it depends on a series of external and internal factors. In zebrafish, habituation to the environment is known to have a great effect on behavior. When entering an unknown environment, zebrafish are known to show nervous behavior, detectable from rapid and abrupt movement. It has been shown that this behavior is often linked to higher levels of stress.

Genetic factors may also play a role in the reaction to a novel environment. When comparing a group of four genetically modified [1] adult zebrafish with a control group of heterozygote animals (Figure 1.1a a, b), we see that both mutants and controls move at greater velocity, and show greater acceleration at the beginning of a 30 minute experiment than compared to later times. There are two main differences between the two groups however: Mutants both start at a smaller speed and acceleration and reach their baseline speed and

[1] The modification affects the MeCP2 gene, which in humans is related to Rett syndrome (RTT) Pietri et al. [32].

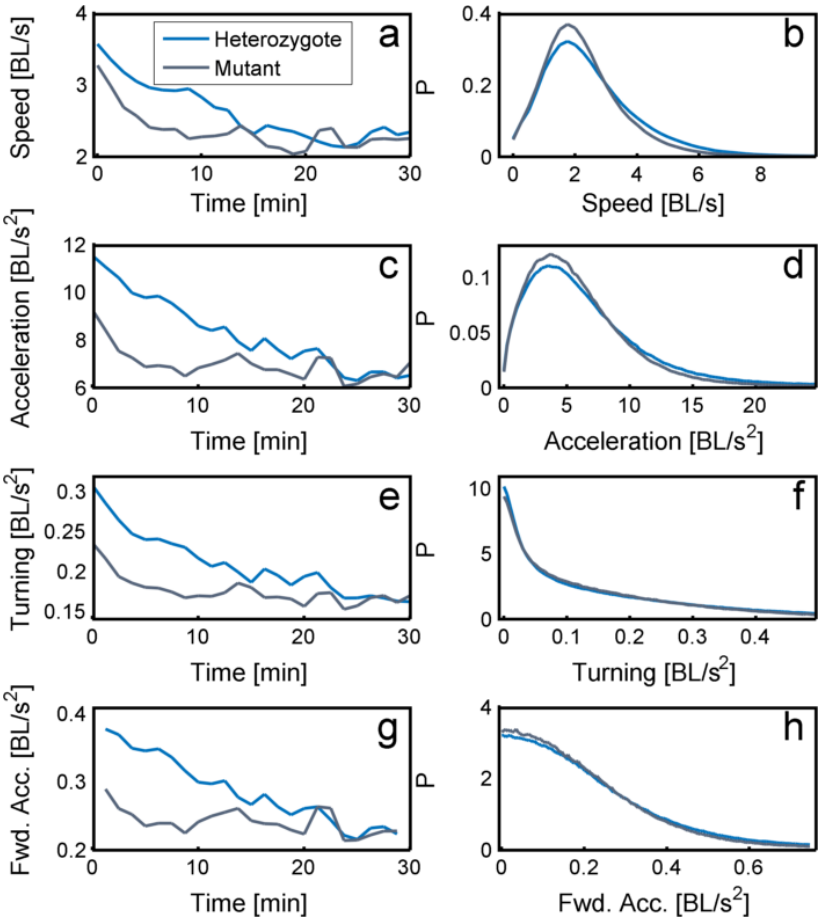

Fig. 1.1 a) Speed and b) Acceleration of mutant MeCP2 zebra fish and a heterozygote control group during the course of a 30 minute experiment.

acceleration much earlier, after approximately 5 minutes as compared to heterozygotes, which need almost 20 minutes to reach the level of MeCP2 mutants.

Speed and acceleration of movement provide an indicator for behavioral changes and a tool to quantify and compare different behaviors, like the effect of habituation and genetics which we have seen for MecP2 zebrafish. A possible interpretation of the observed differences might involve physiological differences in the way mutants and heterozygote controls cope with stress.

Approach Time

Complementary to studying the turning and acceleration behavior of an individual separately, we can analyze the time an individual needs to get from position 1 at $\vec{r_{T_0}} = (x_{T_0}, y_{T_0})$ at time

$t = T_0$ to position 2 at $\vec{r_{T_1}} = (x_{T_1}, y_{T_1})$ at time $t = T_1$, which will include both the time the individual needs to turn and the time it needs to accelerate and approach the new position.

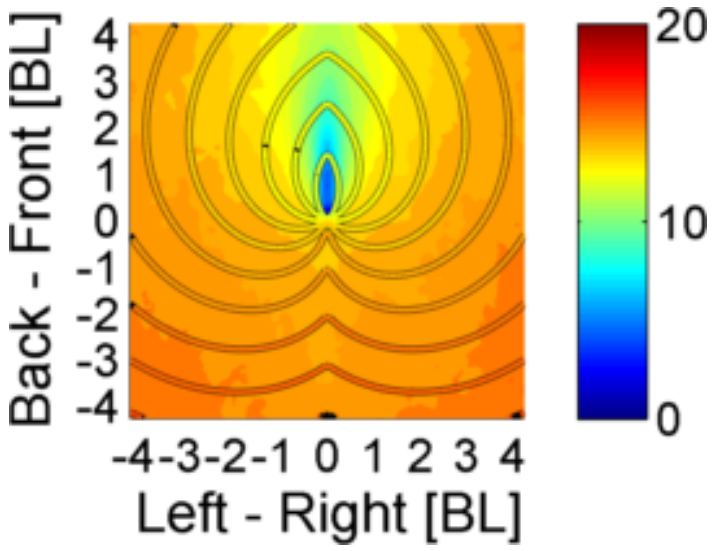

Fig. 1.2 Approach time: Time in seconds an individual needs to get from position (0,0) to any position (x,y). Contour lines depict the result of fitting Eq. 1.1 to the data ($A = 0.53 s/BL$, $B = 1.76 s/rad$ and $C = 10.19 s$).

Figure 1.2 displays the results for an individual in a group of 5 zebrafish. The focal individual is located at the origin. The vertical axis corresponds to the direction of movement of the individual at $T = T_0$. Color indicates the average time the individual needed to move from the origin to the specific position.

A good approximation for the result found in Figure 1.2 is given by a formula based on Krause and Tegeder [21],

$$T(d,a) = A \cdot d + B \cdot a + C. \tag{1.1}$$

where d is the distance between the two positions, a is the angle between the orientation of the individual at position 1 and the vector pointing from the individual to position 2. A, B and C are parameters which can be interpreted as a time offset (C) with units $1\,s$, B a characteristic turning speed of units $1\,s/rad$, and A a characteristic speed of units $1\,s/BL$

1.3 Social Aggregation: Schooling and Shoaling

In the previous section, the study of individual speed and acceleration was sufficient to detect the effect of habituation on zebrafish behavior (Figure 1.1a, b). Habituation however can also have a great effect on the social behavior within a group. In an unknown environment or when feeling threatened, zebrafish are known to form tighter groups which provide better protection from predators and facilitate the exploration of the surroundings. The degree of cohesion of a group can be studied by measuring the distances between the members of the group.

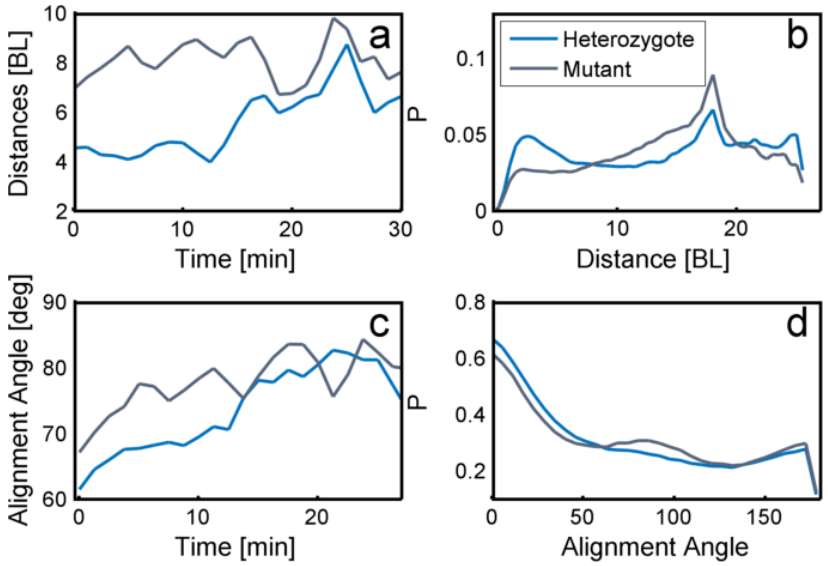

Fig. 1.3 a) Inter-Individual Distance and b) Alignment Angle for mutant MeP2 zebra fish and a heterozygote control group during the course of a 30 minute experiment.

In Figure 1.1c), the heterozygote control group increases inter-individual distances after habituation, when they are familiar with their environment and feel safe from external threats (see also 1.5). The average inter-individual distance is about $4\,BL$, and increases to $7\,BL$ after about 20 minutes, in accordance with the changes in speed and acceleration seen in Figure 1.1a) and b). In comparison, the mutants, which we have seen to show less reaction to habituation, have an inter-individual distance of about $8\,BL$ throughout the experiment.

The first collective phenomenon which comes to mind when dealing with fish most certainly is the synchronized and highly coordinated swimming of a group of fish, known as schooling. Schooling is thought to provide fish an advantage when it comes to predator avoidance, foraging of patchy resources (Grünbaum and Gru [15]) and reducing the energy

necessary for traveling. Common characteristics of animal schools are cohesion, the polarity of the group when individuals align their body orientation, and the spatial and hierarchical structure of the school.

Aggregations of fish are highly adaptive in their behavior and adjust to their ecological context. Thus, a group of fish may change its behavior from schooling to a state of lesser synchronization known as shoal, for example when the threat of a predator is over or the group arrived at a food source. Moreover, different species show more or less tendency to form schools.

In the following, we present exemplary analyses and comparison between schooling and shoaling behavior, represented by a group of free swimming medaka fish (schooling) and zebrafish (shoaling).

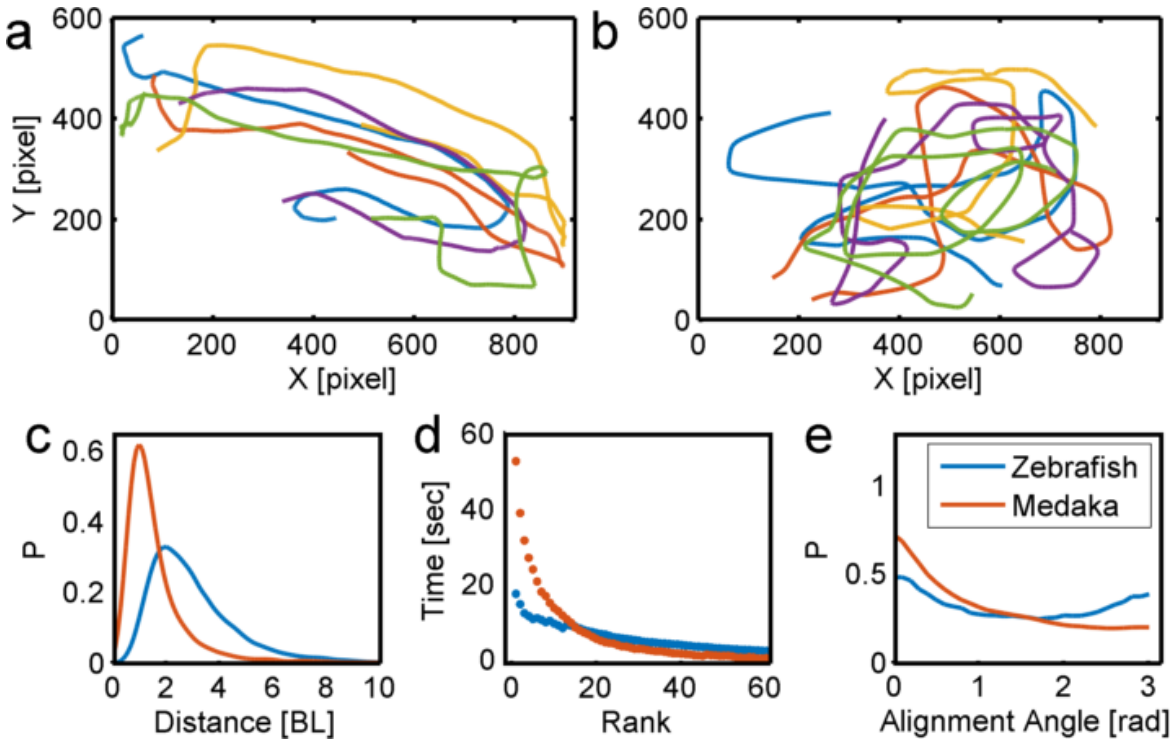

Fig. 1.4 Schooling in Medaka and Shoaling Behavior in zebrafish. a) 10s-Example trajectory of 5 medaka fish b) 10s-Example trajectory of 5 zebrafish c) Distribution of inter-individual distances d) Duration of periods spent within a radius of 3 BL from a conspecific e) Angle between the direction of movement of two animals, averaged over the whole group.

A mere look at the trajectories of animals moving in a group gives us an idea of the synchronicity of their movements. Figure 1.4 compares the movements of a group of medaka

with those of a group of zebrafish. While trajectories of medaka (Figure 1.4a) are well aligned, trajectories of shoaling zebrafish do not seem too well organized (Figure 1.4b).

In order to quantify this first impression, we apply two basic measures to the trajectories of the respective species. The first is the inter-individual distance between the animals of a group. Figure 1.4c shows the probability of finding the nearest neighbor of an individual at a certain distance. Apart from the fact that for zebrafish, the distance which has highest probability lies at about $2BL$, and for medaka at only $1BL$, speaking for a higher cohesion in Medaka, the actual comparison between schooling and shoaling behavior concerns the width of the two distributions: Shoaling zebrafish shows a greater variability in their distance to the nearest neighbor than medaka, compatible with the fact that also the positions of individuals in the group are more variable than in medaka.

Similar average inter-individual distances, and even similar distance distributions, can originate from quite different behavioral patterns, since they do not take into account how the distance changes with time. A stationary group of shoaling animals may show a high variability of distances between group members even over short periods of time, oscillating between short and long distances. On the other hand, a group of fish moving together to a common destination might show low inter-individual distances until they reach their destination and switch from schooling to shoaling behavior, resulting in a sudden change in inter-individual distances.

The stability of social interactions over time can be assessed by looking at the duration of uninterrupted periods of time which animals spent close to a conspecific. Figure 1.4d shows the 60 longest periods of time any pair of medaka or zebrafish spent within a mutual distance of $3\,BL$ from each other. While the longest interaction between two medaka lasted for almost one minute, zebrafish only reached a maximum duration of about 20 seconds. Thus, pairs and groups of medaka show greater temporal stability than those of zebrafish. Also see Chapter 2 for an application of this method.

The greater cohesion present in schooling groups goes hand in hand with a synchronization of the direction of movement of individuals. By assessing the angle between the directions of movement of individuals, we see that the tendency to align is much more pronounced in medaka, for which being in perfect alignment with its neighbor is far more probable than for shoaling zebrafish, as can be seen by the maximum at an angle of $0\,rad$. Zebrafish however show a second preferred angle at π or $180\,deg$, corresponding to an anti-parallel alignment.

Schooling hierarchies

The high polarity and organization of a group of schooling fish leads to the question about the stability of the spatial and hierarchical structure of the group. For many animal species, studies have shown that individuals take their position within the moving group due to their hierarchical status (Nagy et al. [25]). For schooling medaka fish, the hierarchy is stable over days, and the leaders which decide the direction of travel of the whole group can be found predominantly at the front of the group (Pérez-Escudero et al. [30]). In Figure 1.5, the hierarchy of a group of 10 medaka fish can be seen for a 30 min experiment. Numerated circles in Figure 1.5a-c represent the individuals of the group, and the vertical position reflects the rank an animal holds within the moving group when it comes to taking decisions about the direction of movement of the group. The individual on top of the graph is the overall leader. For each pairwise relationship, an arrow points from the leader to the follower.

Even though the shown hierarchies are not completely stable over time, changes are not random, as suggested by Figure 1.5d. Here, the linear ranks of individuals are shown, obtained by ordering individuals corresponding to the vertical position they occupy in the hierarchies. For the first 15 minutes of the experiment, only few individuals change their relative rank. Pérez-Escudero et al. [30] demonstrated that these differences in rank can be significantly smaller than one would expect for a random order of the group.

If one member of a group of animals decides the direction of movement of the whole group and initiates changes of direction, in general this leader will be detectable as the first individual heading in a direction which its followers are copying with a short delay. By measuring this delay in the pairwise correlation of movement direction between animals, the hierarchical order in the moving group can be deduced (Nagy et al. [25], Pérez-Escudero et al. [30]). The position of an individual in the hierarchy is calculated as the mean over the delays it has with respect to all of its neighbors, and the leader of the group is the individual with the smallest negative mean delay of all group members.

Mutual positions

The structure and dynamics of an animal group is determined by the spatial preferences of each of its members. How an individual does react to the presence of a conspecific not only depends on the distance, but also on the relative position and orientation of the two individuals.

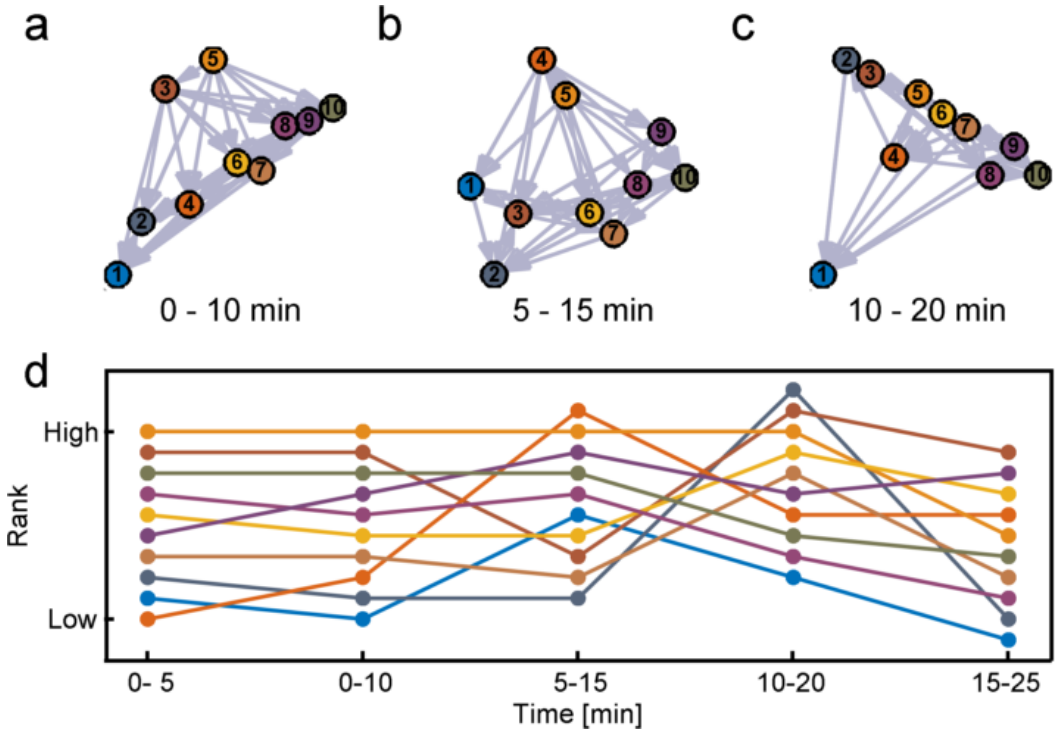

Fig. 1.5 a) - c) Hierarchies obtained from the correlation delay in the directions of movement for a group of 10 medaka fish in various time segments during a 30 minute experiment. Each numbered circle represents one individual of the group. b) Individual relative ranks versus time. Each circle corresponds to the individual of the same color as in a)-c).

In order to keep coordinated with the group, an individual may not only try to maintain a certain distance from its neighbor, but also may react to the relative position and orientation of its neighbors.

Finding the spatial preferences of each member of a group can reveal information about individual differences and the hierarchical role they play within the social structure of the group. Moreover, by studying average preferences of a whole group of fish, we can deduce fundamental characteristics underlying the mechanisms necessary for the coordinated movement of the group.

The spatial preferences of an individual can be measured as the probability of finding a neighbor at a certain position in the individual's natural coordinate system in which the origin coincides with its current position, and the vertical axis with its direction of movement. The frequency of a certain spatial configuration relative to the focal individual at the origin can then be obtained by counting the frames for which the neighbor can be found at a finite region of the focal's coordinate system.

1.4 Shoaling and Rules of Interaction

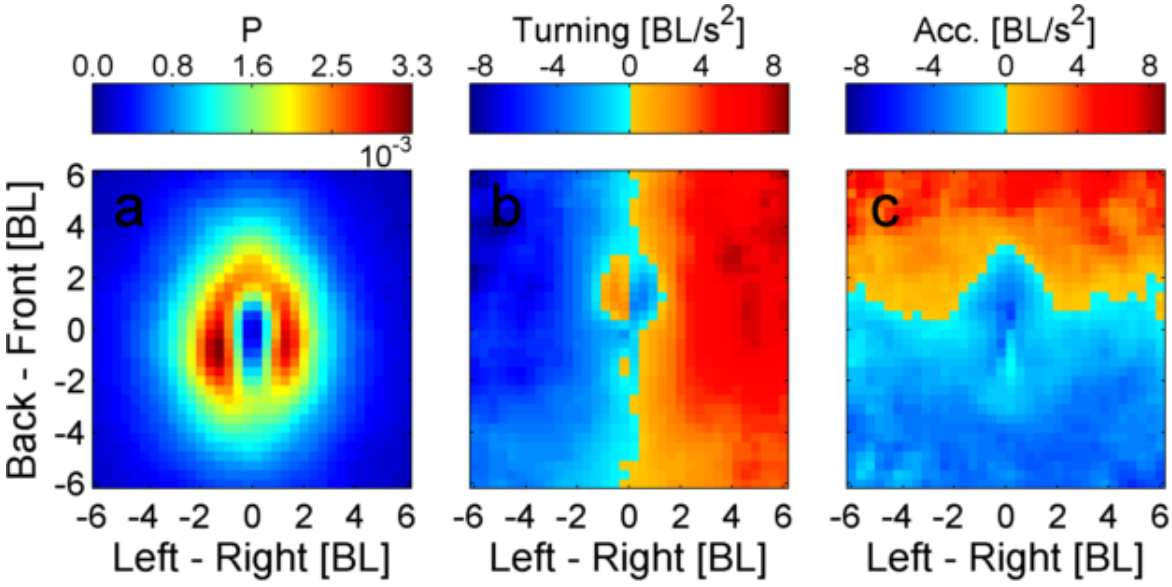

Fig. 1.6 a) Probability of finding a conspecific at a given position in the natural coordinate system of the focal. b) Average turning strength and c) average acceleration strength of the focal as reaction to a conspecific at a given position.

Even in non-synchronized swimming behavior in social animal groups there must be mechanisms by which the cohesion of the group is guaranteed. Even before scientists had at their disposal the experimental possibilities they have today, rules of interaction were hypothesized which might explain the observed collective phenomena. These hypothetical rules of interactions have been used in computer simulations in order to model the aggregation and schooling behavior of animal groups. Usually, these models state a set of rules of interaction in order to determine how an individual reacts to its neighbors in terms of attraction towards, repulsion from, and, in some models, alignment relative to its neighbors.

Some of these features have been found experimentally in different species of fish (golden shiner *Notemigonus crysoleucas* Katz et al. [20]), mosquitofish [16]). Attraction and repulsion can be detected by measuring the change of velocity and direction of an individual in response to the presence of a neighbor: An individual which perceives a nearby neighbor will adjust its direction by turning towards or away from the neighbor, and will increase its speed or slow down in order to approach or avoid the neighbor.

In Figure 1.6 we present the average reaction of zebrafish to a neighbor depending on the relative position. The data is obtained from three experiments with pairs of free-swimming

animals. The results are comparable to those found for golden shiners and mosquitofish: Animals are attracted towards their neighbors when they are further away, and avoid their neighbors when they come too close. Figure 1.6a shows in detail the strength of turns depending on the neighbors' relative position. Red/warm (blue/cold) colors represent rightward (leftward) turns, coordinates in the plane specify the relative position of the neighbor in the natural coordinate system of the focal individual, in which the focal is located at the origin and the vertical axis points in the direction of movement of the focal. The focal individual turns away from a neighbor in front when it is closer than approximately $3BL$, and towards the neighbor when it is further away. In addition, the focal individual mostly accelerates towards a neighbor in front, but slows down when it is behind or at close range, less than approximately $3BL$, in front of the focal (Figure 1.6b).

In Chapter 2, instead of the average magnitude of acceleration and turns towards a neighbor, the probability of these behaviors is used in order to study changes in dynamic interaction during the development of zebrafish larvae.

1.5 Aggression, Dominance and Territoriality

Thanks to its extensive behavioral repertoire, zebrafish not only serves as a model organism for collective behavior, but is also used for the study of aggressive and dominant behavior (Oliveira et al. [27], Dahlbom et al. [11]). Zebrafish is known to form dominance hierarchies in which the dominant animal is determined by aggressive fights between individuals (Oliveira et al. [27]). This agonistic behavior is a good example for the adaptation of behavior to the ecological context: Aggressive behavior is hardly observed in groups of zebrafish in a novel, unknown environment in which the priority is exploration and alertness towards possible threats, for which cooperative behavior is advantageous. In an environment perceived as familiar and safe from external threats however, priorities shift to behaviors linked to foraging and mating, for which a position high up in the dominance hierarchy presents an advantage over competitors.

Aggressive behavior involves individuals chasing one another, characterized by fast swimming and abrupt acceleration of the aggressor towards the chased individual (Kalueff et al. [19]), many times followed by bites. Thus, analyzing movement data for individuals whose speed exceeds a lower limit above the average speed, while heading towards a conspecific, produces a measure for chasing and aggression. Figure 1.7b shows changes in aggressive

behavior in a group of five zebrafish. While during the first 20 minutes a relatively high ratio of chases can be observed, the ratio decreases during the course of the 150min of the experiment.
Figure 1.7 d, f and h show the intensity with which each individual is involved in chasing behavior in the three periods of time shown in Figure 1.7 a. Each bar presents one individual, positive values correspond to a higher number of active chases, while negative values mean that an individual has received more attacks than it has launched itself.

At the same time the dominance hierarchy is established, territories form within the arena. While at the beginning of the trial, all individuals move freely around the whole arena (Figure 1.7c), after one hour individuals already start separating and occupying different areas of the setup (Figure 1.7e). After 2 hours, clear territories can be distinguished (Figure 1.7g). A basic measure of the degree of separation of the territories can be obtained by calculating the average overlap between the territories (Figure 1.7b), which is close to 1 at the start of the experiment, corresponding to an almost complete overlap, but is smaller than 0.4 at the point when the territories are fully established.

The probability of finding an animal in a certain region of the habitat as shown in Figure 1.7 correlates with the territory of the respective animal. Solid borders mark regions in which an animal spent 90% of the time.

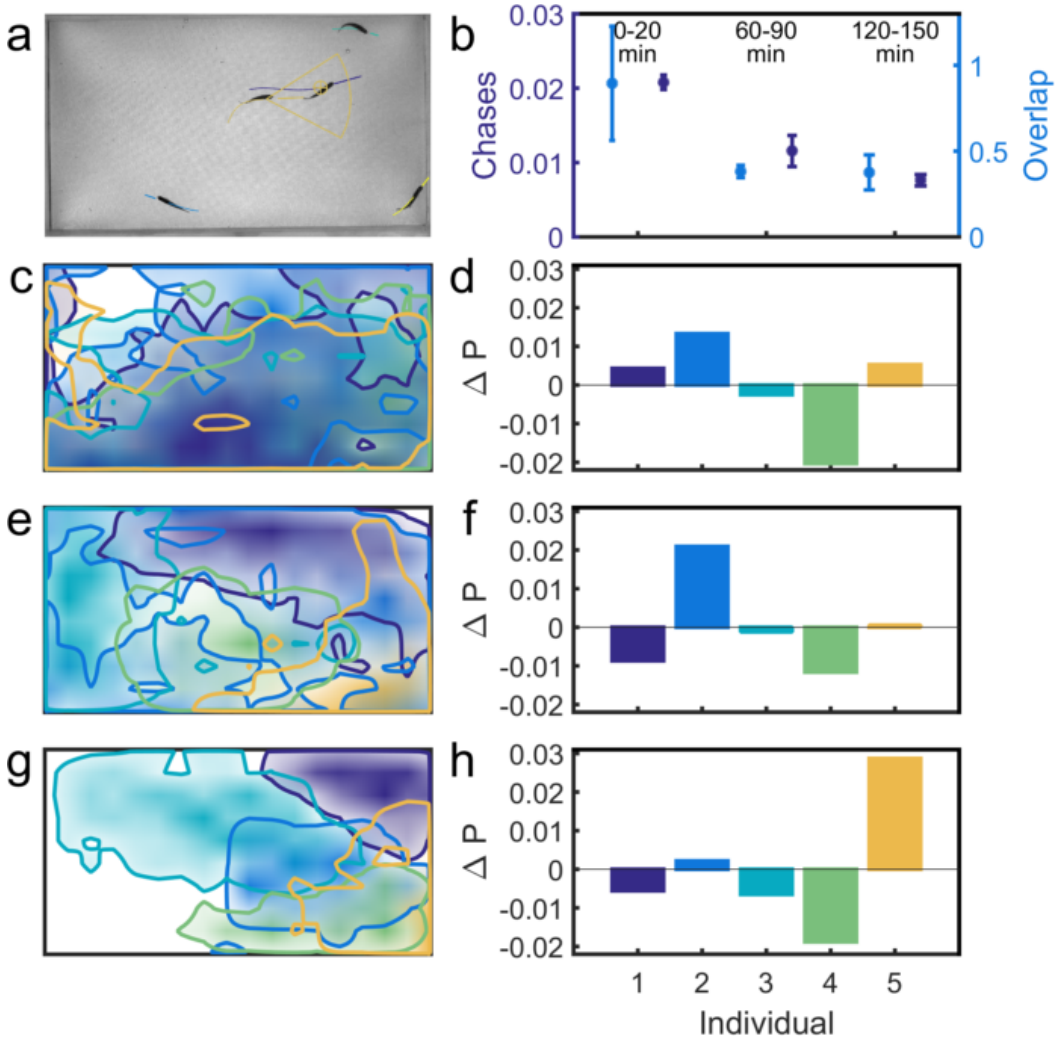

Fig. 1.7 *Aggression and Territoriality*: a) Formation of territories. Probability of finding an individual at a certain position of the arena during 10 min periods a) at the start, c) after 1h and e) after 2h of the experiment. Higher opacity represents higher probability. Lines encircle the region where the individual spent 90% of its time. b),d) and f) show the difference of the frequency with which an individual has been found chasing a conspecific and the frequency of being chased for the corresponding periods. Each color in a-f) represents one individual. g) Average normalized overlap between the territories in a), c) and e). h) Overall probability of chasing behavior.

1.6 Conclusions

- *idSocial* is a Matlab toolbox for the analysis of animal behavior, especially social behavior in zebrafish. It contains a collection of methods for the analysis of basic locomotor behaviors, social aggregation and shoaling, schooling and schooling hierarchies, and aggression, dominance hierarchies and territoriality.
- Methods in *idSocial* base their analysis on spatial coordinates of animals measured with a high temporal resolution, usually extracted from video recordings. In connection with the tracking system *idTracker*, *idSocial* is able to measure the behavior of each individual of a group separately.
- Locomotor behaviors are analyzed in terms of speed, acceleration, propulsion and turning behavior.
- Methods for the analysis of social aggregation, shoaling behavior and schooling look at inter-individual distances and the relative orientation between group members.
- Rules of interaction in social behavior can be inferred from measuring the acceleration of an individual depending on the relative position of the conspecific.
- Hierarchies in schooling are obtained from delay times which occur when individuals copy the direction of movement of conspecifics.
- Aggression and dominance hierarchies can be deduced by measuring the number of chases. Chasing behavior is defined in terms of inter-individual distance, relative orientation and speed.
- Territorial behavior in zebrafish is analyzed by looking at the spatial distribution of individuals in an experimental arena during a period of time. The overlap between territories of different individuals gives a simple measure of the stability of territories.
- New types of analysis can be included easily in *idSocial*.
- *idSocial* offers a framework for the analysis of big data sets, with data from several experimental trials. Trials can be divided into subgroups (e.g, experimental and control groups) and subgroups (e.g., days).

1.7 Conclusiones

- *idSocial* es una colección de herramientas de Matlab para el análisis del comportamiento de animales, especialmente el comportamiento social de pez cebra. Contiene una serie de métodos para el análisis de comportamientos locomotor de locomoción básicos, de agrupación social ('shoaling'), de movimientos coordinados en grupo ('schooling'), de jerarquías en el movimiento coordinado en grupos, agresión, jerarquías de dominación y territorialidad.

- La base para los métodos de *idSocial* la forma el análisis de coordenadas espaciales de animales, obtenidos con alta resolución temporal, en general extraídos de grabaciones de vídeo. En combinación con el software *idTracker*, *idScial* es capaz de medir el comportamiento de cada individuo de un grupo por separado.

- *idSocial* analiza comportamientos locomotor mediante la velocidad, la aceleración total, propulsión (la aceleración paralela a la dirección del movimiento) y giros (la aceleración perpendicular a la dirección del movimiento)

- Los métodos para el análisis de la agrupación social ('shoaling') y el movimiento coordinado en grupo ('schooling') miden la distancia entre los individuos y la orientación relativa entre los miembros de un grupo.

- Se pueden inferir reglas de interacción en el comportamiento social analizando la aceleración de un individuo en función de la posición relativa de un congénere.

- Las jerarquías en el movimiento coordinado de un grupo se obtienen mediante los tiempos de retraso con los que los individuos copian la dirección del movimiento de sus congéneres.

- La agresión y jerarquías de dominación se pueden deducir midiendo el número de persecuciones entre individuos. Las persecuciones se definen mediante la distancia entre los individuos, la orientación relativa y la velocidad.

- El comportamiento territorial en pez cebra se analiza midiendo la distribución de los individuos en la arena experimental durante un periodo de tiempo. El solapamiento entre territorios de animales distintos ofrece una medida básica de la estabilidad de los territorios.

- Se puede añadir fácilmente nuevos tipos de análisis a *idSocial*.
- *idSocial* ofrece un marco para el análisis de grandes conjuntos de datos, que pueden contener datos de varios experimentos. Permite la división de datos en grupos (por ejemplo grupo experimental y control) y subgrupos (por ejemplo para experimentos correspondientes a diferentes días).

1.8 Materials and Methods

In this section, we give a theoretical description of the methods used in this chapter. For more examples, see Chapter Emergence of Social Behavior in Zebrafish Larvae.

1.8.1 Velocity and Acceleration

The velocity vector of an individual at time t is calculated from its trajectory as the difference between its x-y-coordinate at time t and time $t-1$,

$$\vec{v}(t) = (x(t), y(t)) - (x(t-1), y(t-1)). \tag{1.2}$$

Accordingly, acceleration is calculated as the difference between the velocity at time t and $t-1$,

$$\vec{a}(t) = \vec{v}(t) - \vec{v}(t-1). \tag{1.3}$$

1.8.2 Transformation to the Focal System

Those types of analysis which involve the components of acceleration parallel ("forward acceleration","deceleration") and perpendicular ("turning") to the direction of movement of an individual are carried out in the coordinate system defined by the position of the individual (origin) and its direction of movements (vertical axis). Hence, the individual under consideration is called the "focal individual" or "focal", and its conspecifics "neighbors". The direction of movement in frame t is calculated from the normalized velocity vector

$$\vec{n} = |\vec{v}(t)| = \begin{pmatrix} x \\ y \end{pmatrix}. \tag{1.4}$$

In order to transform all neighbor coordinates to the focal system, first the angle between the focal's direction of movement and the vertical axis of the global stationary coordinate

system is calculated as

$$\gamma = \begin{cases} \arctan\frac{y}{x} & \text{if} x > 0 \\ \arctan\frac{y}{x} + \pi & \text{if} x < 0, y \geq 0 \\ \arctan\frac{y}{x} - \pi & \text{if} x < 0, y < 0 \\ \frac{\pi}{2} & \text{if} x = 0, y > 0 \\ -\frac{\pi}{2} & \text{if} x = 0, y < 0 \\ 0 & \text{if} x = 0, y = 0 \end{cases} \tag{1.5}$$

which returns an angle in the range of $[-\pi, \pi]$.
If $\vec{r_f}$ and $\vec{r_n}$ are the coordinates of the focal and the neighbor in the global coordinate system, and the vector pointing from the focal to the neighbor is given by $\vec{\rho} = \vec{r_n} - \vec{r_f}$, the position of the neighbor in the focal system is given by the transformation

$$\vec{\rho_T} = \mu \cdot \vec{\rho} \tag{1.6}$$

where μ the rotation matrix

$$\mu = \begin{bmatrix} \cos\gamma & -\sin\gamma \\ \sin\gamma & \cos\gamma \end{bmatrix} \tag{1.7}$$

1.8.3 Turning and Forward Acceleration

The strength of turns, t, and forward acceleration, f, of an individual can be determined from the acceleration vector $\vec{a}$ as defined in Eq. 1.3 as the component perpendicular and parallel to the direction of movement, or equivalently, as the x- and y-coordinate of the acceleration vector transformed to the focal system, as introduced in Section 1.8.2,

$$\vec{a_T} = \mu \cdot \vec{a} = \begin{pmatrix} t \\ f \end{pmatrix}. \tag{1.8}$$

1.8.4 Approach Time

The approach time Δt is the time an individual needs to get from its position $r(t)$ at time t to a position $r(t + \Delta t)$. The map of average approach time shown in Figure 1.2 is obtained by determining the position $r_T(t + \Delta t)$ at time $t + \Delta t$ of the individual in the natural coordinate

system of the focal defined at time t by the direction of movement $\vec{n}(t)$ and the absolute position $r(t)$ of the individual (as explained in Section 1.8.2).
For a series of moments t of the experiment, the calculation of $r(t+\Delta t)$ is repeated for a number of time steps Δt, with $\Delta t \in (t \quad T]$, where T is the maximum approach time which will be taken into account. In order to calculate the average approach time for a given position, the coordinate system of the focal is divided into bins of fixed width centered at coordinates $c_{ij} = (x_i, y_j), \quad i, j = 1..N$, where N is the number of bins in x- and y-direction. Finally, the mean of all approach times falling into a certain bin is calculated.

1.8.5 Inter-Individual Distance

Inter-Individual Distance between two individuals is calculated for each frame as the euclidean distance between the x-y-coordinates of the focal,(x_f, y_f), and the neighbor individual, (x_n, y_n),

$$d = \sqrt{(x_f - x_n) + (y_f - y_n)}. \tag{1.9}$$

1.8.6 Alignment Angle

The angle between movement directions of two animals as shown in Figures 1.3c and d and 1.4e is calculated from the vectors pointing in the direction of movement of two animals, $\vec{n}_i = |\vec{v}_i|, \quad i = 1, 2$, as

$$\phi = \arccos(\vec{n_1} \cdot \vec{n_2}). \tag{1.10}$$

ϕ lies in the range $[0, \pi]$. See Chapter 2.4 for an additional example.

1.8.7 Duration of Interaction

The duration of an uninterrupted period of time spent close to a conspecific is defined as the time ΔT during which the distance between the two animals has been constantly smaller than a chosen limit D,

$$\Delta T = T_1 - T_0, \quad d(t) < D \quad \forall \quad t \in [T_0, T_1], \tag{1.11}$$

where $d(t)$ is the inter-individual distance as defined in Eq. 1.9.

1.8.8 Correlation Delay

Follower-leader relationships are obtained from the direction of movement $\vec{n}_1(t)$ and $\vec{n}_2(t+\Delta t)$ of the two animals (Eq. 1.4) by calculating the scalar product for different delay times Δt and choosing as the final delay ΔT the value for which the result of the scalar product is maximized, corresponding to a minimization of the angle between $\vec{n}_1(t)$ and $\vec{n}_2(t+\Delta t)$. In other words, if ΔT is positive, at time $t+\Delta t$ individual 2 will be heading in the direction individual 1 was moving at time t, and thus individual 2 is following individual 1.

1.8.9 Territories and Territory Overlap

Territories are obtained by dividing the arena into bins of fixed width, centered at coordinates $c_{i,j} = (x_i, y_k), \quad i = 1..N_x$ and $j = 1..N_y$, where $N_{x,y}$ is the number of bins in x- and y-direction. Then the number of frames $F_{i,k}$ for which an individual can be found at the position of bin $c_{i,k}$ is measured for a number of frames F_{total}, and the result for each bin is normalized,

$$f_{i,k} = F_{i,k}/F_{total}. \tag{1.12}$$

Finally, a territory is defined as the area where an individual has spent a total of 90% of the time of the experiment, corresponding to a number of $0.9 \cdot F_{total}$ frames.

The overlap of territories belonging to different individuals A and B is calculated by

$$U = \sum_{i}^{N_x} \sum_{k}^{N_y} \sqrt{f_{i,k}^A} \cdot \sqrt{f_{i,k}^B} \tag{1.13}$$

1.8.10 Approach Angle

The approach angle is the angle between the orientation vector $\vec{n}$, given by the direction of movement of the focal, and the vector $\vec{\rho}$ pointing from the focal to the position of the neighbor:

$$\theta = \arccos(\vec{n} \cdot \vec{r}). \tag{1.14}$$

Here, θ lies in the range $[0, \pi]$. If θ is positive (negative), the neighbor is located at the right (left) hand side of the focal. Although no example of the approach angle has been presented in this chapter, it is used to detected chasing behavior, see the next section. Also see Chapter 2.3 for an example.

1.8.11 Chases

Chasing behavior can be detected by looking at the speed and direction of movement of an individual: The movement of the focal individual is considered a chase, if

- the distance to the conspecific is smaller than a chosen maximum, $d < R$,
- its speed exceeds a given limit, $|\vec{v}| > S$, and
- the magnitude of approach angle (see Sec. 1.8.10), is smaller than a limiting angle, $\theta < \Theta$

An example is given in Figure 1.7.

1.8.12 Relative Positions

In the focal system defined in Section 1.8.2, the neighbor position is given by the vector pointing from the position of the focal at the origin to the transformed coordinate of the neighbor.
The orientation of the coordinate system is chosen to be clockwise, such that a positive (negative) perpendicular component of acceleration corresponds to a right (left) turn. A positive parallel component means forward acceleration, while a negative component can mean both deceleration and backward acceleration. Maps of relative neighbor positions, turning, acceleration/propulsion and attraction are generated dividing the space around the focal in rectangular bins of fixed width. In Chapter 2, polar maps were created by dividing the plane in bins defined by angular edges at $[0, \pi/4, \pi/2, \ldots, 2\pi]$ and radial edges at $[0, 1.5, 3, \ldots, 6]BL$.
For maps of the probability of finding a neighbor at a certain area of the coordinate system of the focal, the number of frames for which the neighbor was found within the corresponding bin was normalized by the total number of frames and by the size of the bins, given by

$$A = \alpha \cdot dr(r + dr/2), \tag{1.15}$$

where α is the angular width of the bins, r is the distance of the inner edge of the bin from the center, and dr is the radial width of the bins.

1.8.13 Maps of Turning, Acceleration and Attraction Strength and Probability

Force maps show the average reaction of an individual depending on the relative position of a conspecific. The reaction can be measured as turning towards or away from the conspecific, or an increase or decrease in speed (see Section 1.8.3).
In Chapter 2, in addition to turning and forward acceleration, *attraction* is introduced as a measure of social attraction towards a conspecific: Positive attraction is present when the magnitude of the angle between the vector of acceleration and the vector connecting the position of the focal with the position of the neighbor is less than 90 degrees.

To generate the respective force map, for turning, acceleration/deceleration and attraction the respective magnitude is calculated for the focal as described in Sections 1.8.1, 1.8.2, and for each frame is sorted into the bin corresponding to the neighbor position in that frame, as introduced in Section 1.8.12.

Finally, either the probability or mean or median strength of the respective behavior can be calculated. In order to obtain the probability of the respective reaction, the number of frames corresponding to a right turn (for turning), forward acceleration (for acceleration/propulsion) or positive attraction (for attraction) in each bin is divided by the total number of frames in that bin. For the mean or median, the respective operation is applied to the data in each bin.

Chapter 2

Emergence of Social Behavior in Zebrafish Larvae

2.1 Introduction

New tracking techniques, methods of analysis and mathematical modeling are allowing a quantitative characterization of the interaction rules underlying group behavior (Katz et al. [20], Herbert-Read et al. [16], Arganda et al. [3], Pérez-Escudero et al. [30]). An additional level of analysis would include how brains process information and make decisions. Zebrafish (*Danio rerio*) is a especially suitable species showing group behavior and it is possible to image the brain dynamics at cellular resolution (Ahrens et al. [1], Ahrens et al. [2], Panier et al. [28], Naumann et al. [26], Portugues et al. [33]) , perform targeted mutations (Hwang et al. [18]) and manipulate circuit dynamics by genetically expressing light-activated channels (Arrenberg et al. [4]). Imaging and optogenetic methods are best suited for early larval stages so the genetic expression of activity and manipulation probes is maximal and light for imaging of activity and for circuit manipulation pass through the transparent skin.

However, research on interactions among zebrafish has given contradictory results. The first studies reported that this animal is only attracted to others at 3 weeks (Engeszer et al. [13]), an age in which neurobiological techniques have very limited application. A later study reported attraction at 6-7 days post fertilization (dpf) (Hinz et al. [17]), but recent studies have only found attraction at 2 weeks using very large number of animals to find statistical significance (Dreosti et al. [12]). A common feature of these studies is that they measure attraction by looking at the proportion of time a single individual spends at the side

of a set-up with several conspecifics instead of a region with none. Conspecifics are always separated from the individual by a transparent wall. This separation limits the interactions to the visual system, impeding natural interactions which also involve sensory information from the lateral line and the olfactory system. In addition, results can be affected by the preference for walls and corners (thigmotaxis) present in zebrafish. A study conducted using an open field task with free-swimming zebrafish (Buske and Gerlai [8]) found inter-individual distances smaller than expected from random controls at an age as early as 7 dpf, but findings may have been influenced by additional, non-social environmental stimuli.

To be able to measure quantitatively rich interactions, we developed idTracker (Pérez-Escudero et al. [30]), a tracking system of individuals in groups with no error propagation, obtaining large high quality data sets. We reasoned that a quantitative measurement of rich interactions would trace the onset of attraction behavior in zebrafish larvae and the details of which and how its characteristics change in development. We studied interactions from 6 dpf to 24 dpf in an experimental arena of 19 BL diameter (adjusting the absolute diameter between 7.5 to 13.5 cm, depending on bodylength) and approximately 0.5 to 1.5 cm water depth (Figure 2.11b, see also Section 2.4.2).

2.2 Results

2.2.1 Inter-Individual Distance and Relative Positions

We first studied interacting pairs to measure their relative configuration in space. We took each individual as a focal animal at the center of a coordinate system. We then measured the position of the second fish in all frames of the video with respect to the focal animal (Figure 2.1A, focal and fish in blue and orange, respectively). We then computed the probability of finding the second fish in the coordinate system of the focal. We performed the average of this probability for all focal fish we had for each age (Figure 2.1B). The example corresponds to 24 dpf and it is clear that the second fish has a high probability to be close to the focal, larger at the sides. We also obtained a randomized control using as the position of the second 'virtual fish' that of the fish at a random point of the video sufficiently separated in time from the focal (12000 frames, corresponding to about 6.6 min in our experiments) to avoid the influence of actual interactions (Figure 2.1C). The randomized control also has a maximum at the origin of the coordinate system as a result of how fish visit their environment with a preference for the boundary (a model with both fish randomly distributed in the arena

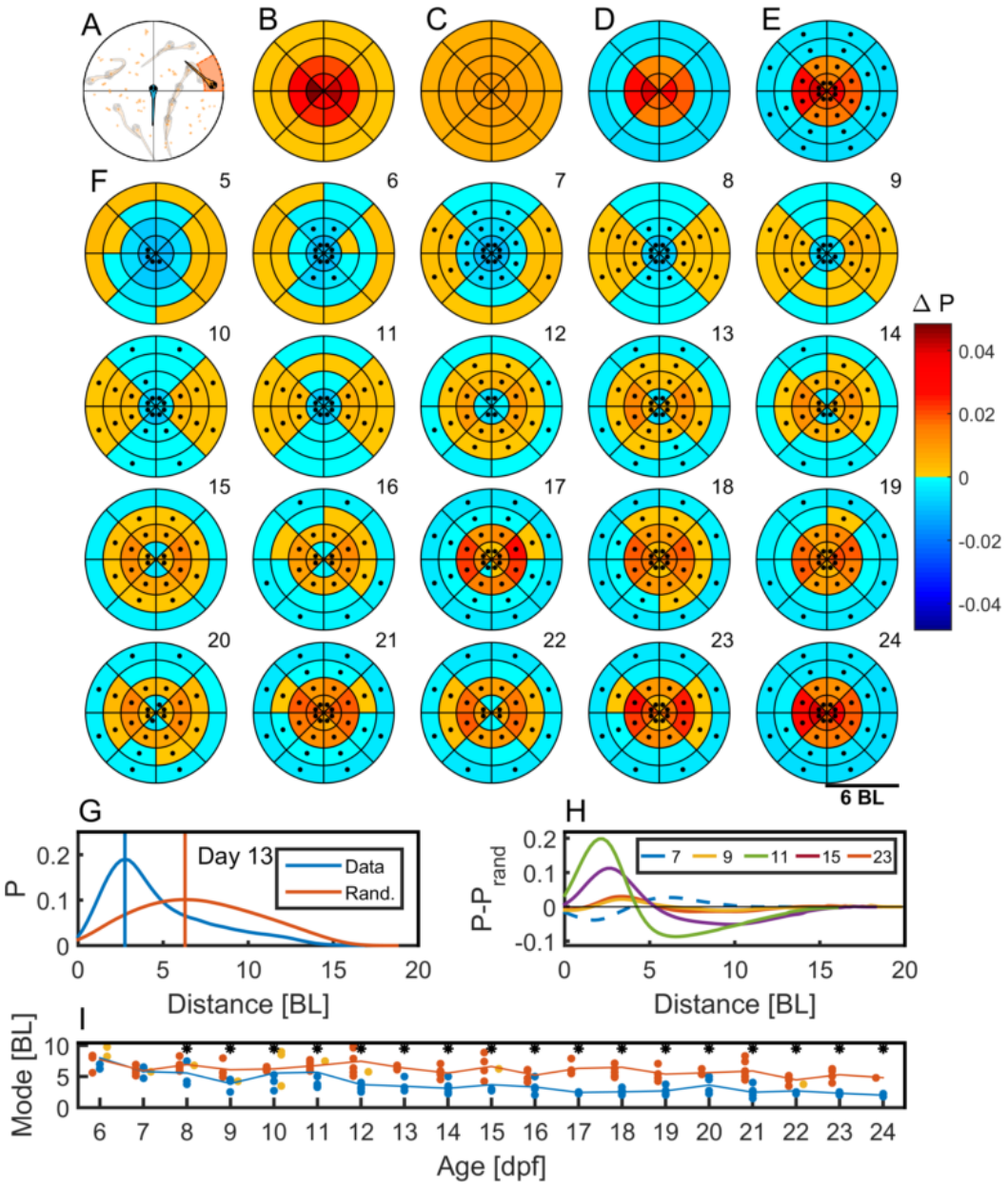

Fig. 2.1 *Inter-Individual Distance and Relative Positions* A) Natural coordinate system of the focal individual (blue) used to determine the probability of relative positions of the neighbor individual (orange) B) Average probability of spatial configurations for zebrafish of age 24 dpf. C) Corresponding probability from randomized control data. D) Difference of the probabilities shown in B) and C). E) Dots mark sectors which are significantly different from randomized controls (Monte Carlo, $p<0.05$). F) Results obtained by the procedure explained in B)-E) for age 5 to 24 dpf. G) Example of the distribution of inter-individual distances and randomized controls for a pair of zebrafish of age 13 dpf. Vertical lines mark the respective mode of the distribution. H) Difference between average distance distributions and controls for various days (Solid lines: Distribution of modes is significantly different from randomized controls. Monte Carlo, $p<0.05$). I) Mode of distance distributions, average over trials (blue solid line, stars indicate statistical significance) and for each pair of individuals (blue dots: Significantly different from randomized controls (Monte Carlo, $p<0.05$), yellow dots: No significant difference). Orange dots/lines represent results from randomized controls.

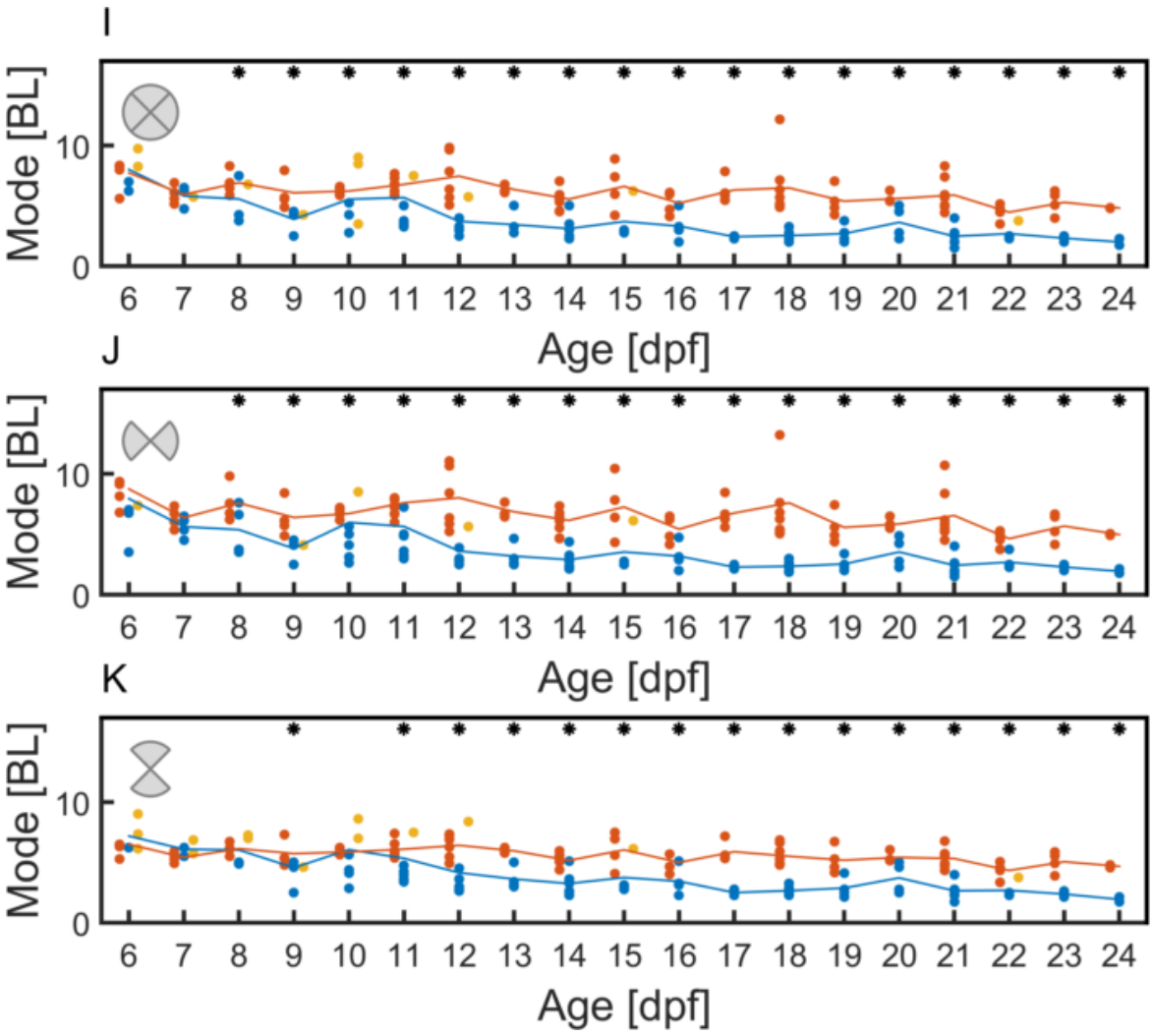

Fig. 2.2 *Inter-Individual Distance.* Mode of distance distributions, averaged over all pairs of the same age (blue solid line, stars indicate statistical significance) and for each pair of individuals (blue dots: Significantly different from randomized controls (Monte Carlo, $p<0.05$), yellow dots: No significant difference), I) taking into account all neighbors, J) only neighbors on the left/right, and K) only at the front/back. Orange dots/lines represent results from randomized controls.

obtains a similar distribution, see Figure 2.13C). To clearly show the difference between the experimental and randomized configurations, we subtracted the two and represented in warm and cold colors those regions in which the experiment gives higher and lower probabilities than the control, respectively (Figure 2.1D). We then ran a significance analysis for each sector to test whether the experimental probabilities are different to randomized controls, and added a dot when P<0.05 (Figure 2.1E). For 24 dpf, the second animal can be found in a radius less than 3BL with a significant high probability and more often at its sides than in front-back positions.

We found 4 stages in the development of relative spatial configurations. At 6 dpf there is only a significant region of low probability at close and medium distances (day 6, Figure 2.1E). From 7 to 11 dpf there is a low probability region at very close distances and also at medium distances at the front and back similar to those at day 6, but also regions of probability higher than in the control configuration to the left and right, similar to the ones observed in older animals but of lower values (days 7-11, Figure 2.1E). From day 12 to 14 the front-back region changes from lower to higher values than the randomized configuration (days 12-14, Figure 2.1E). In older animals, the region of repulsion at low distances is dramatically reduced and the vast majority of the regions at close and mid distances have a probability much higher than the random control (days 15-24, Figure 2.1E).

We also measured the distribution of distances between the pair of fish along the video, averaged over at least four pairs of fish of the same age (blue, Figure 2.1G) and compared it to the random one (orange, Figure 2.1G), for example by subtracting them (Figure 2.1H). The single parameter that captures best the difference between the experimental and random distributions is the mode, and it is significantly different from the random one for animals from 8 dpf and older (Figure 2.1I). The significance analysis for the distribution of distances can be done for each pair (Figure 2.1I, blue dots for P<0.05, yellow for P>0.05 and orange for randomized controls) or for each age (star for P<0.05, Figure 2.1I). We also performed a complementary analysis of relative angles between the two fish and found that fish tend to be significantly side by side from 6 dpf (Figure 2.3) and swimming in parallel significantly from 8 dpf (Figure 2.4).

2.2.2 Social Attraction

The analysis of relative spatial configurations, distances and angles gives information about the relative arrangements among fish as a result of attraction dynamics but does not analyze

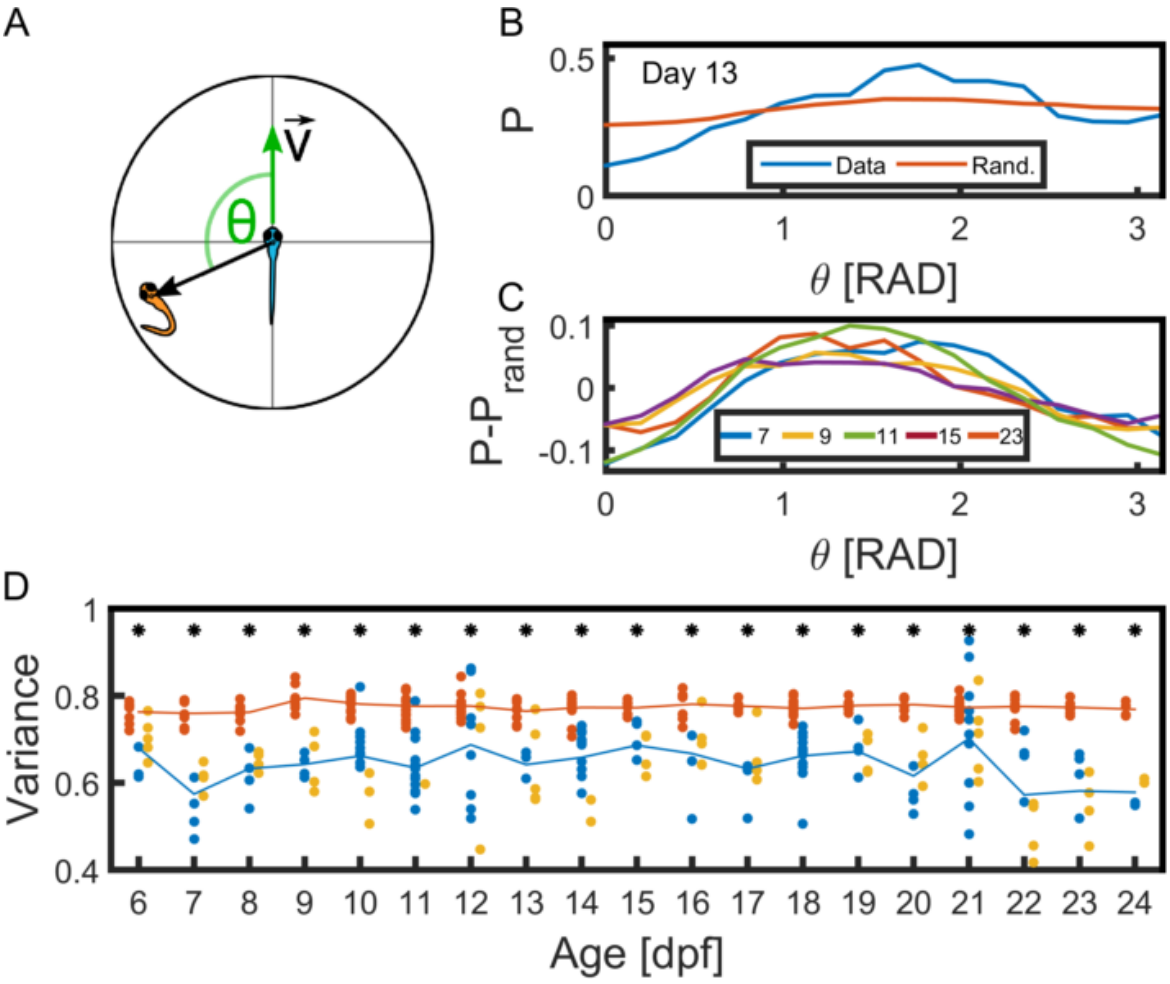

Fig. 2.3 *Relative Angle to Neighbor* defined as the angle θ between the movement direction of the focal and the vector pointing from the position of the focal towards the neighbor (A). B) Example of the distribution of θ and randomized controls for a pair of zebrafish for a pair of age 13 dpf. C) Difference between the average distribution of θ and controls for various days. The variance of the distributions is significantly different from their randomized controls for all ages shown (Monte Carlo, $p<0.05$) D) Variance of the distribution of the relative angle, averaged over all pairs of the same age (blue solid line, stars indicate statistical significance) and for each pair of individuals (blue dots: Significantly different from randomized controls (Monte Carlo, $p<0.05$), yellow dots: No significant difference). Angle between the movement direction of the focal and the movement direction of the neighbor.

attraction directly. A direct analysis of interactions can be done by measuring whether individuals accelerate towards conspecifics. We separated the acceleration of the focal animal into a parallel and a perpendicular component. We analyzed the probability that the perpendicular component points to the right when the second animal is at a given position in space (Figure 2.5A). For example, for day 24, whenever the second animal is to the right

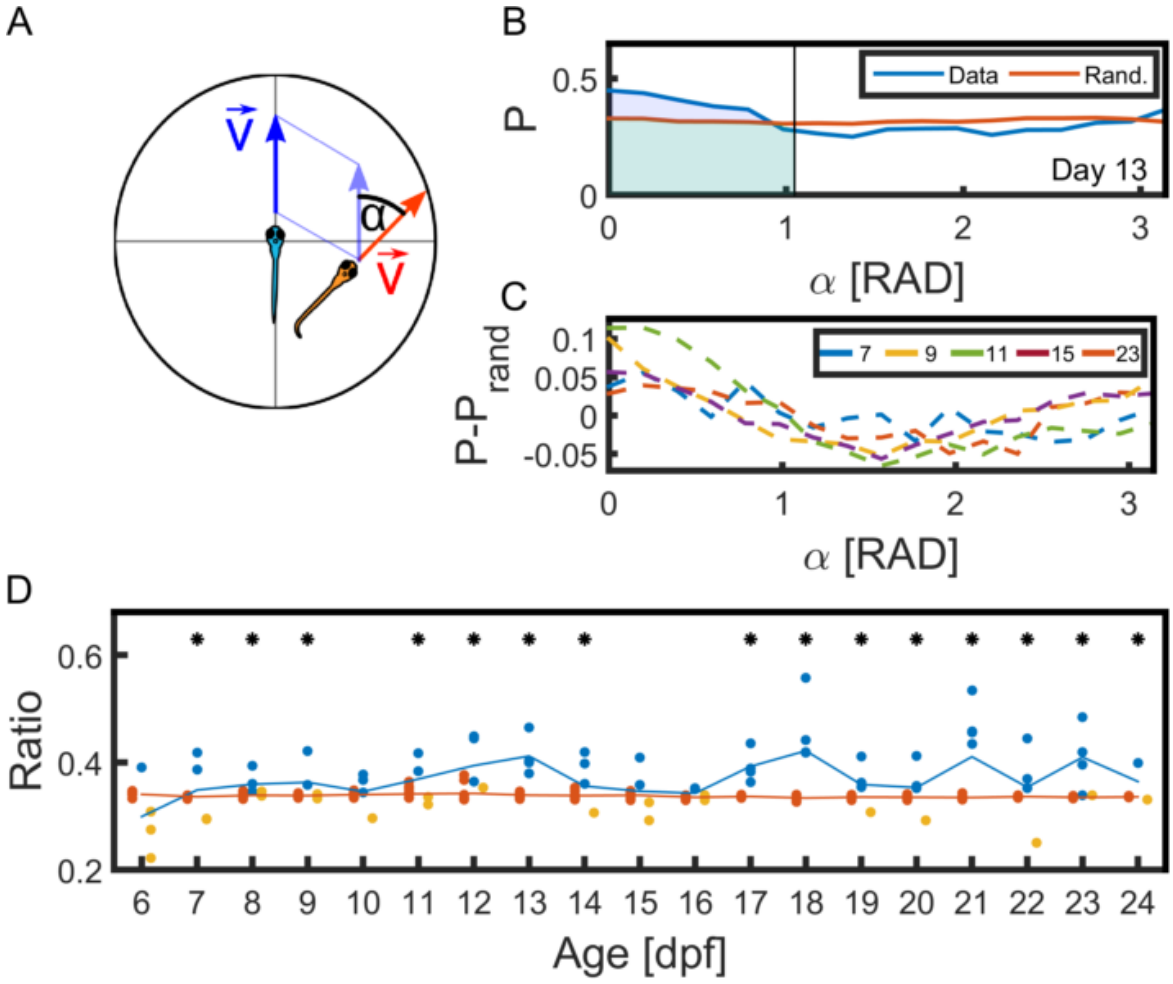

Fig. 2.4 *Relative Angle of Orientation* defined as the angle α between the directions of movement of the focal and the neighbor (A). B) Example of the distribution of α and randomized controls for a pair of zebrafish for a pair of age 13 dpf. The ratio of alignment is calculated as the ratio of frames for which $\alpha < \pi/3$ (indicated by a vertical black line) C) Difference between the average distribution of α and controls for various days. Solid lines indicate that the ratio of alignment is significantly different from randomized controls (Monte Carlo, p<0.05) D) Ratio of alignment, averaged over all pairs of the same age (blue solid line, stars indicate statistical significance) and for each pair of individuals (blue dots: Significantly different from randomized controls (Monte Carlo, p<0.05), yellow dots: No significant difference).

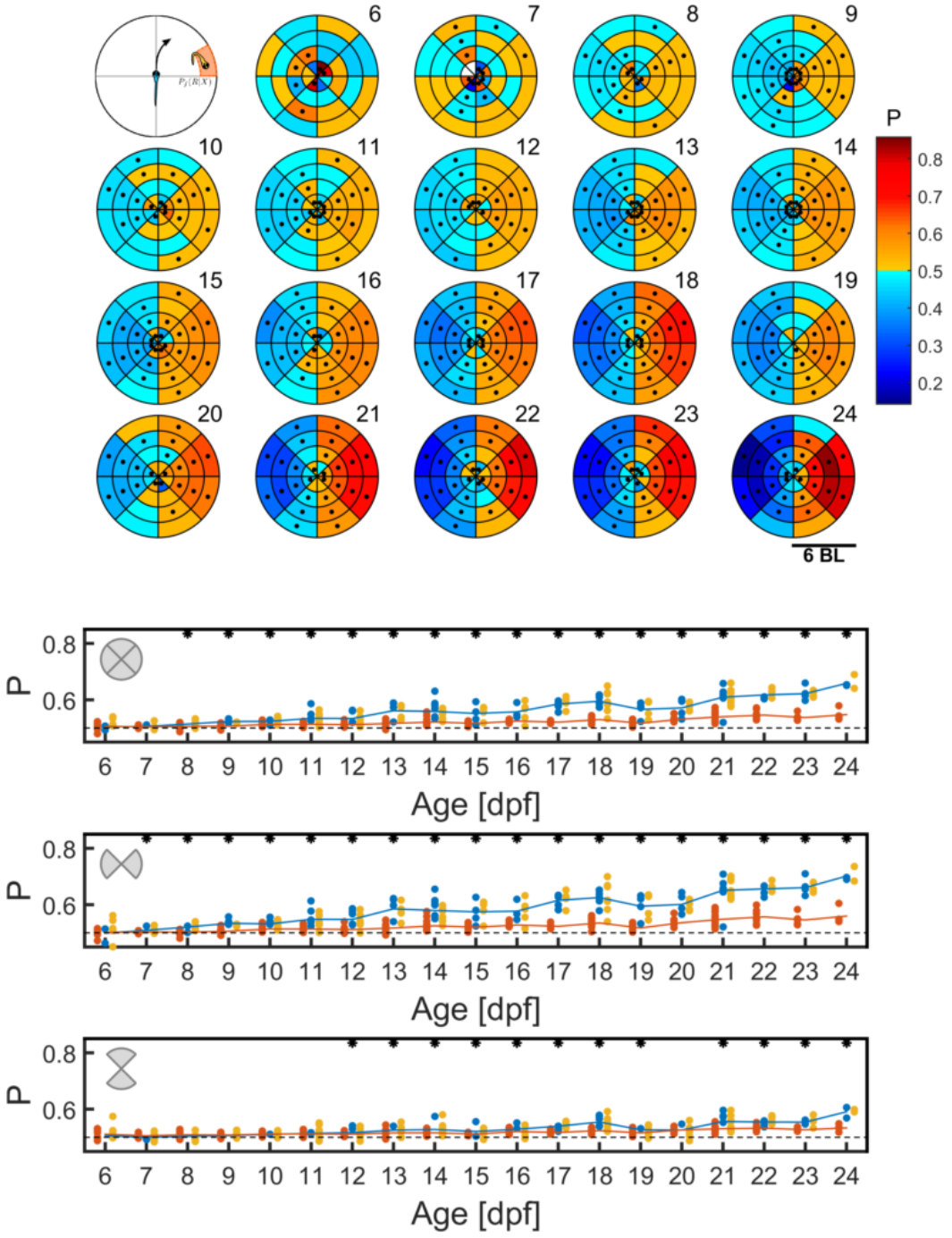

Fig. 2.5 *Turning Behavior as a function of neighbor position.* A) In the coordinate system with the focal at the origin and the vertical axis given by its direction of movement, a probability > 0.5 (< 0.5) of turning right for a given neighbor position is indicated by warm (cold) colors. B) Probability of turning right averaged over all pairs of the same age. C)-E) Probability of the focal turning towards the side of the neighbor, averaged over all pairs of the same age (blue solid line, stars indicate statistical significance) and for each pair of individuals (blue dots: Significantly different from randomized controls (Monte Carlo, $p<0.05$), yellow dots: No significant difference), C) taking into account all neighbors, D) only neighbors on the left/right, and E) only at the front/back. Orange dots/lines represent results from randomized controls.

front-back positions and from 17 to 24 there is a consistent attraction dynamics in all points around the focal (Figure 2.5A).

An analysis of attraction can also be performed without the full spatial information by computing the proportion of frames in which the perpendicular component of the acceleration of the focal animal points to the side in which the other animal is (Figure 2.5B). This measure is significant from 9 dpf and increases with age. The same analysis was done restricted to the cases in which the second animal is to the sides of the focal and significant attraction is then found from 8 dpf (Figure 2.5C). When the second animal is in more front-back positions the left-right attraction is delayed until 14 dpf and with smaller values than when the second animals is to the sides of the focal (Figure 2.5D). While the earliest attraction is found for the group at 8 dpf, a significance analysis at the level of individuals reveals that some of the larvae at 6 and 7 dpf present attraction dynamics (blue dots at 6 and 7 dpf, Figure 2.5B-D).

An analysis similar to that of Figure 2.5 but for the parallel component finds that larvae accelerate towards animals in front of them and decelerate when at the back but its significance starts at 12 dpf (Figure 2.6). We performed an alternative analysis of attraction in space but without dividing the acceleration into perpendicular and parallel components. For this we computed a measure of how the focal accelerates roughly towards the other animal as the probability that the angle between the focal acceleration and the relative position vector between the two animals is less than π. At 24 dpf, this probability is very high (> 0.8) for most points of space in a radius of 6 BL (day 24, Figure 2.7), indicating attraction. At 6-7 dpf we see no consistent attraction, which only starts at 8-10 dpf when animals are side by side (Figure 2.7A). At 11-16 dpf attraction also develops in front-back positions and from 17 dpf gets stronger (Figure 2.7A). The proportion of frames in which the angle of the acceleration of the focal animal is significant at 9 dpf (Figure 2.7B) but at 8 dpf when considering only the sides of the focal animal (Figure 2.7C).

2.2.3 Duration of Interactions

We have seen that larvae can be attracted to each other from 6 dpf, practically from the beginning of behavior, attraction is a common behavior at 8-9 dpf and it matures slowly until 16 dpf and faster after 17 dpf. However, while animals have a significant probability of getting attracted to each other, its value is much lower for the youngest larvae so the times they spend together may be low. As a complementary analysis to understand interactions, we then computed the time two animals spend close to each other in groups of four. Specifically,

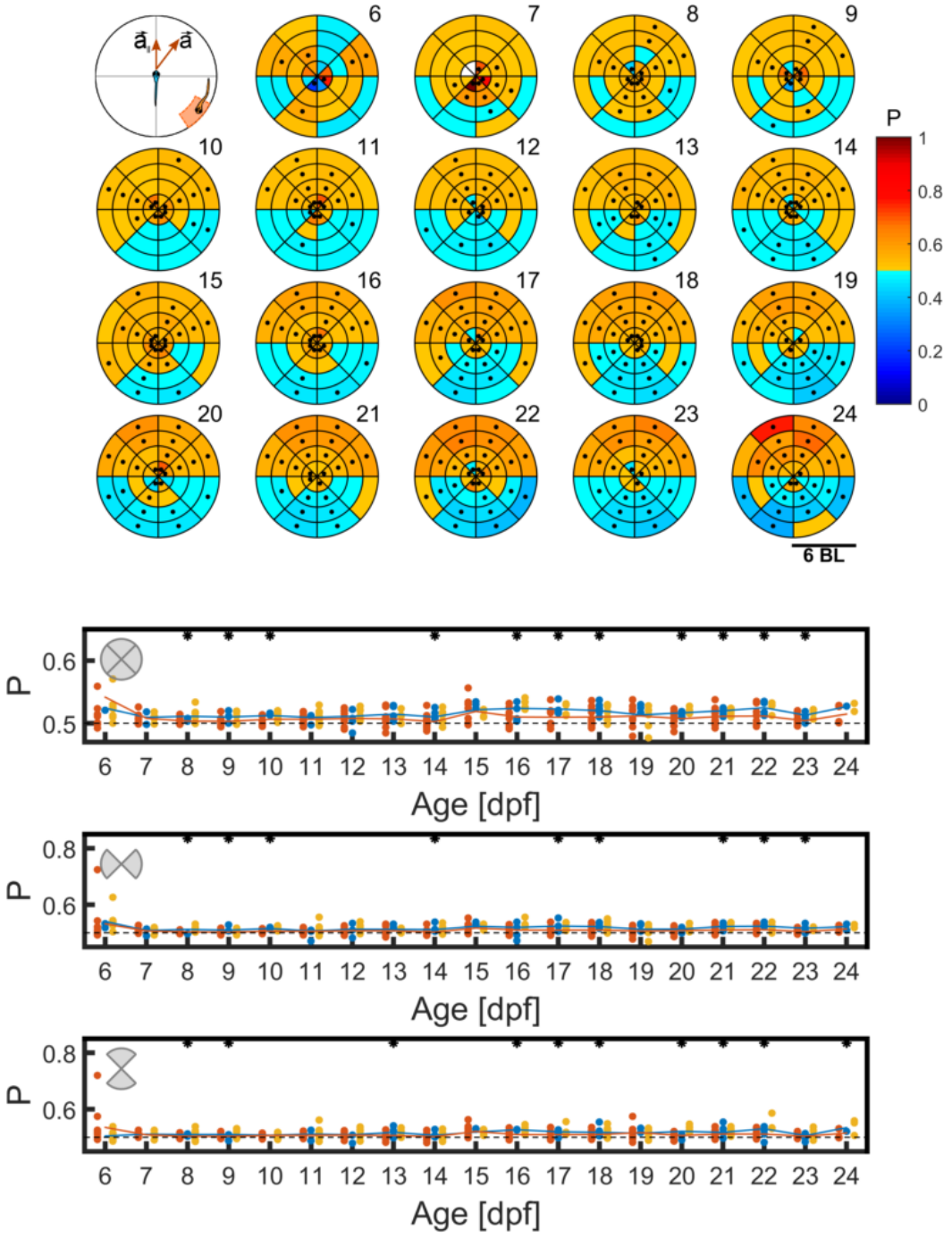

Fig. 2.6 *Acceleration Behavior as a function of neighbor position.* A) In the coordinate system with the focal at the origin and the vertical axis given by its direction of movement, a probability > 0.5 (< 0.5) of forward acceleration for a given neighbor position is indicated by warm (cold) colors. B) Probability of forward acceleration averaged over all pairs of the same age. C)-E) Probability of the focal accelerating/decelerating towards the position of the neighbor, averaged over all pairs of the same age (blue solid line, stars indicate statistical significance) and for each pair of individuals (blue dots: Significantly different from randomized controls (Monte Carlo, $p<0.05$), yellow dots: No significant difference), C) taking into account all neighbors, D) only neighbors on the left/right, and E) only at the front/back. Orange dots/lines represent results from randomized controls.

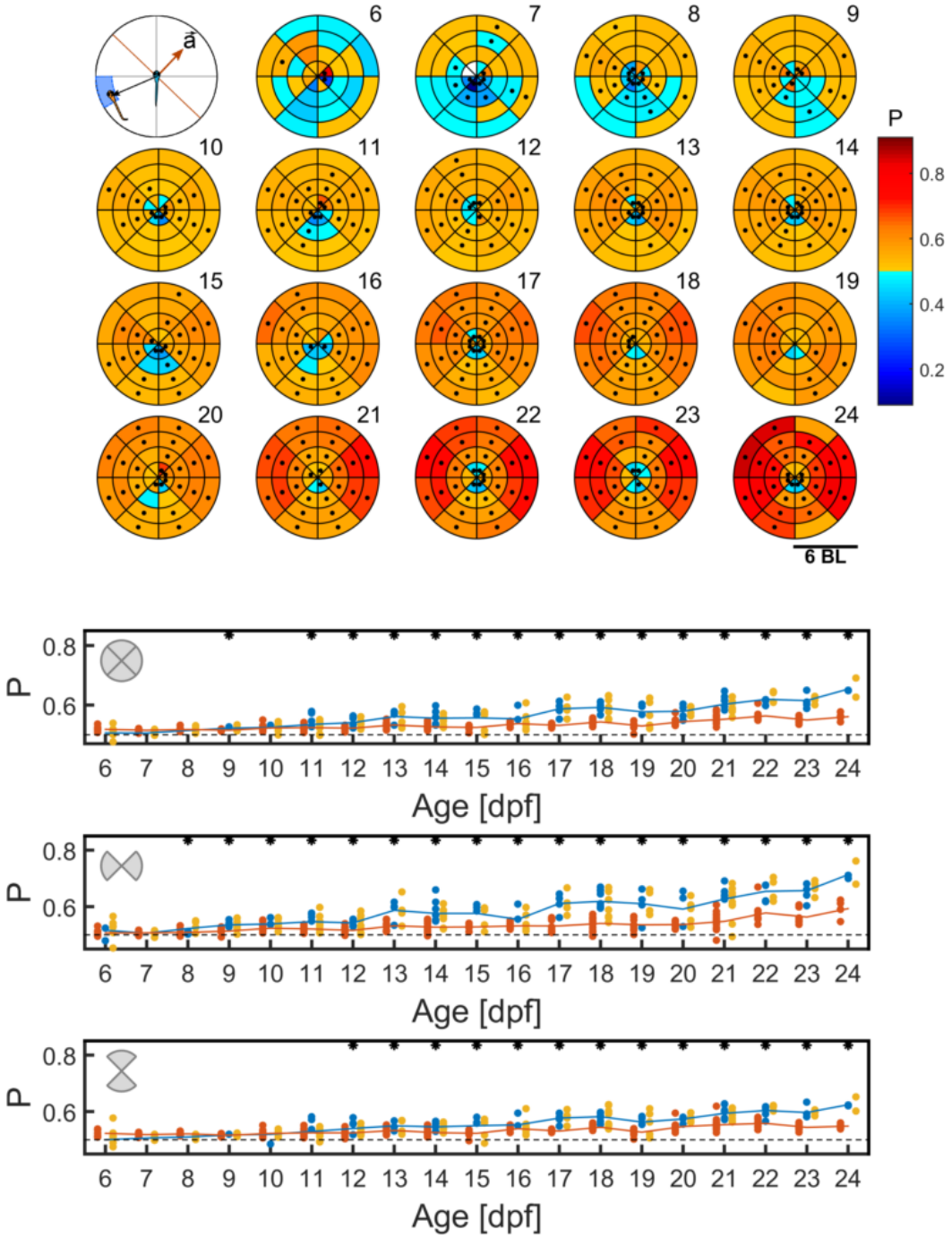

Fig. 2.7 *Attraction as a function of neighbor position.* A) The focal is attracted towards a neighbor if the angle between its vector of acceleration and the position of the neighbor is less than 90 deg. In the coordinate system with the focal at the origin and the vertical axis given by its direction of movement, a probability > 0.5 (< 0.5) of attraction for a given neighbor position is indicated by warm (cold) colors. B) Probability of attraction averaged over all pairs of the same age. C)-E) Probability of attraction towards the position of the neighbor, averaged over all pairs of the same age (blue solid line, stars indicate statistical significance) and for each pair of individuals (blue dots: Significantly different from randomized controls (Monte Carlo, $p<0.05$), yellow dots: No significant difference), C) taking into account all neighbors, D) only neighbors on the left/right, and E) only at the front/back. Orange dots/lines represent results from randomized controls.

we computed the times an animal was continuously at a distance of less than 6 BL from a conspecific and we rank ordered them from largest to lowest time (Figure 2.8). For 21 dpf it is clear that the largest times are much larger than the largest times one can find in the randomized controls (21 dpf, Figure 2.8D). The largest time for 21 dpf is, for example, close to 2.5 minutes in which the animals are continuously close to each other. At 6 dpf, the times of interaction are not different from randomized controls.

For groups of four, we found 5 different stages. In 6-9 dpf no individuals showed any time different from random, in 10 dpf some individuals showed a time slightly different from random but not as an age group, 11-20 dpf are group ages significantly different from the random but with times only about 1.3 times the random ones, while at 21-24 the times are more than twice the random ones. The time animals spend together impact the group sizes we can find at different ages. We computed the probability of finding 1, 2 and 3 conspecifics at less than 6BL of a focal animal (Figure 2.8F). Until 10 dpf the time spent is compatible with random configurations with a decreasing order of 0, 1, 2 and 3 individuals. In 11-18 the number of conspecifics is different from random and the most probable cases are 2 and 1 conspecifics. During this period the probability of having 3 other conspecifics increases and after 19 dpf is the most common configuration.

Using the types of analysis presented above, social behavior can not only be measured for a group as a whole, but also for each animal of a group separately. In Figure 2.9 we present the results for individuals in groups of two for a selection of days. Maps of relative positions, turning and attraction behavior give an overview of the tendency of social interactions. They are completed by the p-values obtained from the comparison between the mode of the distance distribution (see Figure 2.1), the ratio of attractive turning (see Figure 2.5) and attraction (see Figure 2.7) and control data. A score of social interaction is given (see Section 2.4.6 for details). In addition, the distribution of inter-individual distance and the duration of interactions for the focal animal and its conspecific is given.

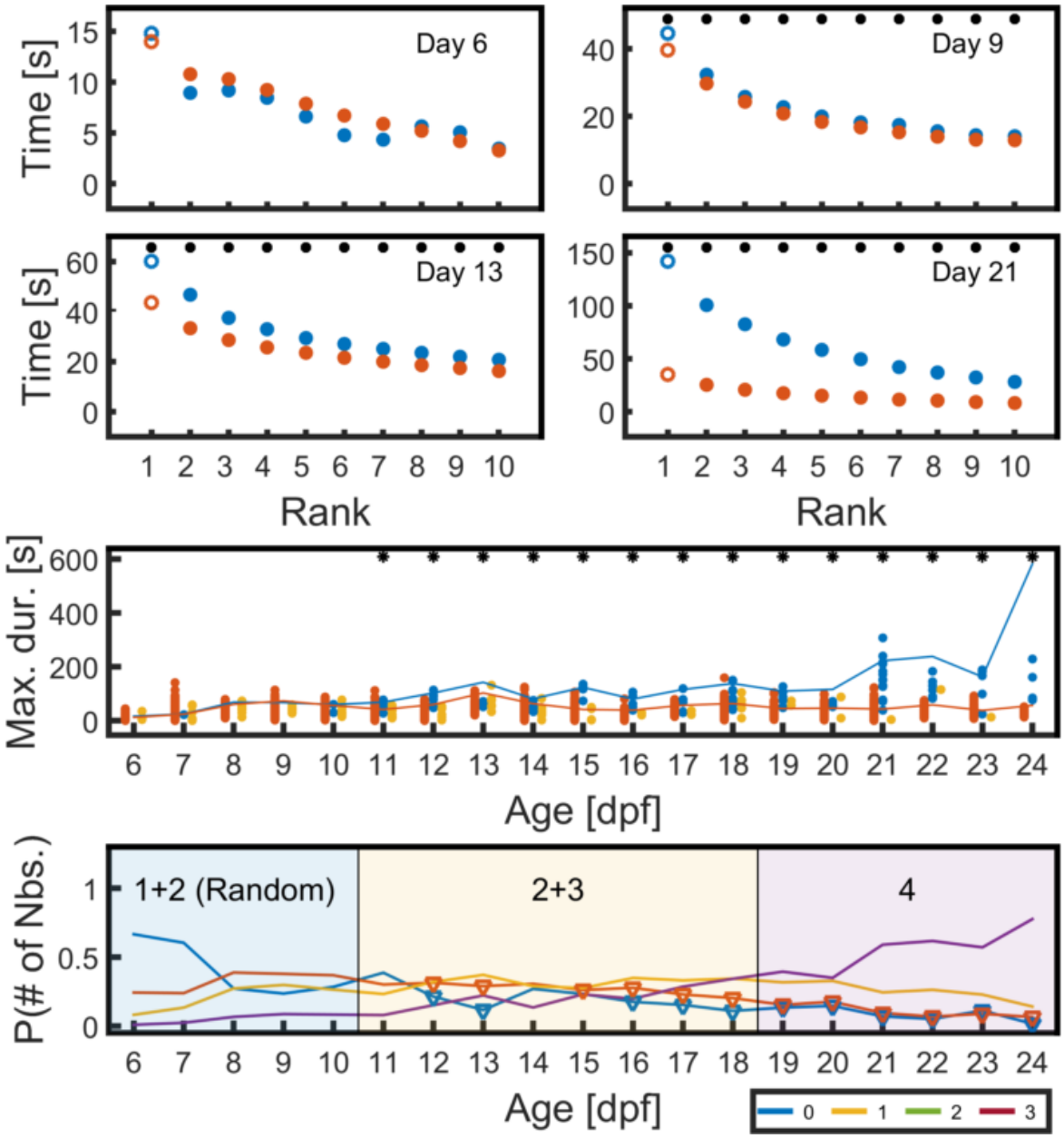

Fig. 2.8 *Duration of Interactions and Group Sizes* A-D) Example "Rank distributions" of duration of uninterrupted periods of time an animal spent within a radius of 6 BL from at least one conspecific in a group of four animals for various days E) Duration of the longest period averaged over all groups of the same age versus age (solid blue line) and randomized controls (solid orange line), and longest times found for each group (blue dots: Significantly different from controls and yellow dots: not significant) and controls (orange dots). F) Probability of an animal being close to a specific number of neighbors. Each colored solid line corresponds to a certain number of neighbors, see legend.

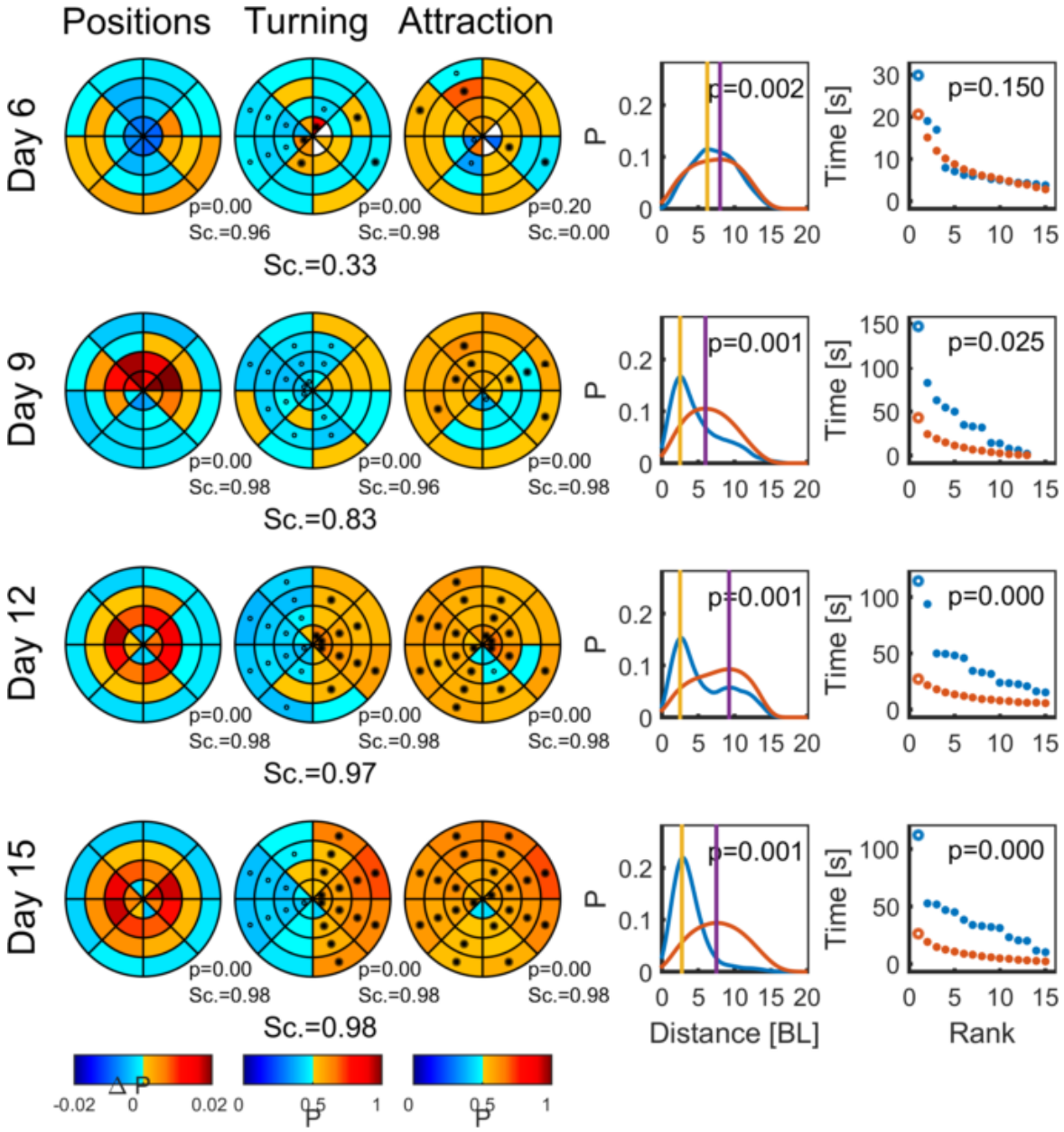

Fig. 2.9 *Analysis of Individuals* From left to right, columns show the map of relative positions, turning and attraction behavior, the distribution of distances and the duration of interaction. Rows correspond to four animals of different age, interacting with one other animal (N=2).

2.3 Discussion

One aim of this study was to proof the applicability and utility of a series of methods to detect and quantify different strengths of social behavior. The first step towards this objective was to verify that *idTracker* can be used for zebrafish larvae starting from an early age. We were able to get good tracking results starting from an age of 5 dpf (see Section B.1; results from day 5 were later excluded from our analysis because animals showed very little movement). Crucial for the successful tracking and behavioral analysis was the careful design and adaptation of the experimental setup, both to minimize external stimuli which could interfere with behavior and to provide adequate optical conditions of the recordings. An additional technical issue concerns the post-processing of coordinates acquired from recordings using *idTracker*. See Sections 2.4.4 and B.3 for an overview of the influence of noise on the analysis and a comparison of different methods for trajectory smoothing.

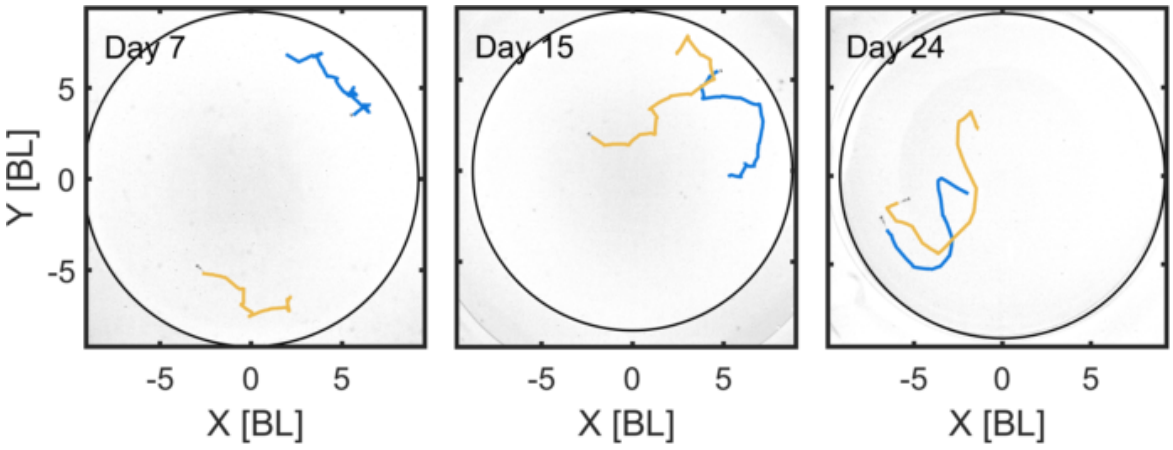

Fig. 2.10 *Example Trajectories* Traces of 7, 15 and 24 day old larvae and juveniles which exemplify the increasingly gregarious behavior of zebrafish with age.

In many cases, methods of analysis similar to those presented here have been used before in other contexts, but not in order to quantify changes in social behavior. Two exceptions here are inter-individual distance and group polarization. Group polarization, comparable to the average relative angle of orientation as shown in Figure 2.4, has been applied by Miller and Gerlai [24] in order to quantify the transition from schooling to shoaling behavior in adult zebrafish. Inter-Individual distance has been used by Buske and Gerlai [8] to measure the development of shoaling behavior with age in zebrafish. Though the latter study shares common features with ours, its focus was rather on the long term development during the first 5 months post fertilization, and did not have at its disposal the possibilities offered by

the automated tracking done by *idTracker*, namely the temporal resolution and duration of the trajectories, which are fundamental for most of the methods presented here.

The principal question of our study was at what age social behavior can be detected in zebrafish larvae. The objective here was to assess the possibility of combining the function of zebrafish as a model organism for genetic and neurobiological research at the one hand and for the study of social behavior on the other. While most of the imaging techniques and opto-genetic tools which make zebrafish the ideal model for studying the nervous system require characteristics like skin transparency, are mainly applicable at early stages of the development, i.e., in embryos and larvae, social behavior in form of aggregation and shoaling cannot be observed until a later stage of development. By determining the moment at which social behavior can be detected in zebrafish, the use of zebrafish as a model in neuro-genetic research of social behavior can be confirmed or ruled out.

Our results show that social behavior can be detected in zebrafish at an age of 8 days post-fertilization on a population level, and even earlier for single individuals. This is an age at which animals are still transparent and opto-genetic and imaging techniques like two-photon microscopy can still be applied. Thanks to the rich data made available by acquiring the full trajectories, it is not only possible to detect social behavior on a population level, but individuals can be assessed separately. Thus, combining behavioral experiments and neurobiological tools, it is now possible to link individual traits of social behavior to the underlying neural mechanisms and processes.

A great concern when measuring social behavior in a laboratory setup is the influence of the geometry of the experimental arena and of external stimuli on individual behavior. All quantities measuring social behavior have to me tested against control data in order to make sure that the measured social interactions are not actually experimental artifacts, generated for example by the preference for the same area of the arena, independently present in all individuals. Possible reasons for such a preference can be heat or light gradients inside the setup, or the preference to stay close to the wall observed in many animal species and known as *thigmotaxis*.

A big effort has been made to minimize these external effects. Firstly, an hour glass was used as experimental arena, lacking actual walls. Thus thigmotaxis decreased (see Section B.2) and shadows at the borders, potentially interfering with both behavior and the quality of the tracking, were eliminated.

In addition, in order to take into account additional unmeasured factors like external environmental stimuli or individual preferences of the animals, we generated control data from randomization of the original trajectories (see Section 2.4.5). This approach, which has not been applied before to the analysis of social behavior in zebrafish, lets us determine the statistical significance of our experimental results.

The results do not only give us the moment of onset of detectable social behavior, they also show clear changes in the way larvae interact during their development. The relative positions shown in Figure 2.1f show a clear shift from a preference for having the conspecific at the side between days 7 to 11 to a more uniform distribution of neighbor positions in a ring around the focal with a radius of about 4.5 BL. Both the origin for the early preference and the later shift are unknown. A possible search for an explanation could look into the role of binocular vision which is known to be used in prey capture in zebrafish larvae (Bianco et al. [5]), and into behavioral lateralization (Miklósi et al. [22]). Interestingly, Dreosti et al. [12] found that one week old zebrafish larvae predominantly orient themselves in a angle of about 90 degrees relative to a stimulus chamber in which they can perceive a group of conspecifics, and shift this preferential angle towards 45 degrees during weeks 2 and 3.

Our results suggest that social behavior develops gradually, starting from the first socially motivated interactions at an age of 7 or 8 dpf, to the formation of stable groups and the observation of shoaling behavior at an age of 21 dpf. In order to form stable groups, zebrafish have to develop a series of abilities. Shoaling and schooling behavior involves a complex interplay between perception ("What does an individual perceive?"), cognition ("Distinguish conspecifics from other stimuli"), and motor abilities ("Are animals quick and skilled enough to navigate efficiently in a group?").

Although we were not able to address these questions directly, we could reveal some crucial aspects of the mechanisms involved in the formation of stable groups. While the measurement of inter-individual distance gives us information about the effect of the mechanisms involved in group behavior, it lacks information about the actual nature of these mechanisms. By studying turning and acceleration behavior however, we are able to quantify the probability of an individual actively interacting with a conspecific, and see that this probability, just like inter-individual distance, increases gradually with age. Specifically, the tendency of turning towards a conspecific, which can be interpreted as the result of perception and cognitive processes which lead to the decision to interact with a conspecific, increases with age, and

together with other mechanisms like the propulsion towards the conspecific eventually leads to smaller inter-individual distances.

By considering only the probability of turns and acceleration towards other members of the group, but not the strength of the acceleration, we lose one aspect of information about the formation of stable groups. This is done deliberately however, in order to focus on the cognitive component of the development of social behavior, and neglecting the influence of the increase of physical strength during development.

Finally, by measuring the duration of uninterrupted periods of interaction as shown in Figure 2.8a-e, we get an estimate of how well animals coordinate their movements in order to stay close to each other, a behavior that requires the continuous active adaptation of the movement of each individual in terms of direction and speed, and thus again involves the active decision of individuals to stay within the group.

Conclusions

- We used a simple experimental arena, consisting in a circular hour glass, to study the development of social behavior in zebrafish *Danio Rerio* larvae of age 6 to 24 days post-fertilization (dpf). The geometry of the hour glass enabled us to adjust the diameter of the arena in order to keep the ratio of bodylength and diameter constant. In addition, the absence of borders improves the visual conditions for the tracking and reduces thigmotaxis.

- With the combination of *idSocial* and *idTracker*, we were able to extract and analyze trajectories from between 4 and 8 20-minute videos of pairs and groups of 4 zebrafish for each day between day 6 and 24.

- In order to study the development and change in strength of social behavior, we analyzed inter-individual distances, mutual attraction, relative orientations, times of interaction between larvae, and the average number of conspecifics within an interaction zone, using methods developed for *idSocial*.

- The statistical significance of the results was determined by comparison with control data which was obtained by randomizing the original data. Thus, the control data takes into account experimental artifacts like the influence of the border of the arena or spatial preferences within the arena.

- We find that the average distance between individuals as measured by the mode of the distance distribution is robustly smaller than controls from day 8 on. Individuals tend to have their conspecific at their side between days 7 and 11, and for later ages in a closed ring around them.

- Zebrafish show robust average turning towards their conspecific from day 8 on, from which moment on turning probability constantly increases until day 24, the last day of measurements. Individuals have a higher probability of social turns for conspecifics at their side in comparison with neighbors in front or behind for age 8 to 10.

- We found that the probability to turn towards a conspecific is the best measure of inter-individual attraction, while the probability of forward acceleration or deceleration depending on the position of the conspecific gives less clear numerical results.

- The ratio of attraction, calculated from the angle between the vecor of acceleration and the angular position of the conspecific detects social behavior starting from day 8 when only taking into account lateral neighbors, and robust attraction from day 10 when not considering the relative position of the neighbor.
- Average times of interaction between members in a group of 4 zebrafish are significantly higher than controls from an age of 9 days onward, with a marked increase from day 15 on.
- Until 10 dpf, the number of conspecifics within a radius of 6 BL is not significantly different from random controls in a group of 4 zebrafish. Between days 11 and 18, the most probable configuration consists in groups of 2 or 3 members, equivalent to 1 or 2 neighbors within the interaction zone. From day 19 onward, a group size of 4 is the most probable configuration.
- While for the whole population robust social behavior was found from an age of 8 dpf, our results suggest that some individuals show increased social attraction and proximity of conspecifics already at earlier ages in comparison to random controls.

Conclusiones

- Utilicé un montaje experimental básico, que consistía en un vidrio de reloj, para estudiar el desarrollo del comportamiento social en larvas de pez cebra *Danio Rerio* entre 6 y 24 días post-fertilización (dpf). La geometría del vidrio de reloj permite ajustar el diámetro de la arena experimental para mantener constante la ratio de la longitud corporal de las larvas y el diámetro. Adicionalmente, la ausencia de paredes verticales mejora las condiciones visuales para la extracción de las trayectorias y reduce la tigmotaxis.

- Combinando *idTracker* y *idSocial*, pude extraer y analizar trayectorias de entre 4 a 8 vídeos de 20 minutos de duración para parejas y grupos de 4 peces cebra para cada día en el periodo de 6 a 24 días post-fertilización.

- Para estudiar el desarrollo y el cambio de la intensidad del comportamiento social, analicé las distancias entre individuos, la atracción social, las posiciones relativas y la duración de las interacciones entre los animales y el número promedio de congéneres dentro de un radio de interacción. Para ello utilicé métodos de *idSocial*.

- Determiné la significancia estadística de los resultados usando datos control obtenidos a través de la randomización de los datos originales. De esta manera, los controles tienen en cuenta artefactos experimentales como la influencia de los bordes de la arena o las preferencias espaciales dentro de la arena.

- Encontré que la distancia promedio entre individuos, medida a través de la moda de la distribución de distancias, es significativamente menor que los controles a partir del día 8. Los individuos tienden a tener a sus congéneres a su lado entre los días 7 y 11, y después en un anillo alrededor.

- En promedio los individuos de pez cebra giran hacía sus congéneres a partir del día 8. A partir de este momento, la probabilidad de girar hacía un congénere aumenta gradualmente hasta el día 24, el último día de experimentos. Entre los días 8 y 10, los individuos tienen una probabilidad más alta de girar hacía congéneres que se encuentran a sus lados con que de girar hacia congéneres situados delante o detrás.

- La probabilidad de girar hacía un congénere es la mejor medida de la atracción entre individuos, mientras que la probabilidad de acelerar o desacelerar en función de la posición de los congéneres proporciona resultados numéricos menos claros.

- El ratio de atracción, calculado a partir del ángulo entre el vector de aceleración y la posición angular del congénere detecta comportamiento social a partir del día 8, si en el análisis solo se incluyen vecinos que se encuentran a los lados, y atracción robusta a partir del día 10 si no se considera la posición relativa del vecino.

- Los tiempos promedio de interacción entre miembros de un grupo de cuatro peces cebra son significativamente más altos que los controles a partir del día 9, con una subida pronunciada a partir del día 15.

- Hasta el día 10, el número de congéneres que se encuentran a una distancia de 6 body lengths (longitud corporal) o menos de un individuo en un grupo de cuatro animales no es significativamente diferente de los controles obtenidos a través de randomización. Entre los días 11 y 18, la configuración más probable consiste en grupos de 2 o 3 miembros, equivalente a un número de 1 o 2 congéneres dentro de la zona de interacción. A partir del día 19, la configuración más probable es la de un tamaño de grupo de cuatro animales.

- Mientras que para la población total se encuentra comportamiento social robusto a partir de 8 dpf, nuestros resultados indican que algunos individuos muestran atracción social significativamente diferente de los controles en edades anteriores.

2.4 Materials and Methods

2.4.1 Housing and Maintenance

Zebrafish embryos of both Wild Type (WT) and AB strains were obtained from ZF Biolabs, Madrid. Until day 5 post-fertilization they where kept in petri dishes of diameter 15 cm inside an incubator with a 14/10 light–dark cycle. The temperature was kept constant at 27.5°C. Larvae were given powder food (sera micron) three times a day once they had started foraging. Excess food and debris was removed in the morning and in the evening. In addition, about one third of the water was replaced with fresh water in the evening. On day 4 dpf, zebrafish larvae were transferred to the experimental setup and kept in fish tanks of dimensions 12x12x30 cm with a weak flow of fresh water. We used two experimental setups with separate closed water circulation systems for WT and AB larvae. Each setup contained two fish tanks with approximately 100 larvae each. During the experiments, only larvae from the same holding tank were grouped together. Both the water temperature and the light–dark cycle were kept the same as in the incubator and were not changed throughout the time of the experiment. The feeding protocol stayed the same as in the incubator until day 12 dpf, when for the midday feeding the powder food was replaced by live artemia. Excess food and debris was removed in the morning before the experiments and in the evening after they had finished.

2.4.2 Experimental Protocol

Experiments for both WT and AB zebrafish were conducted with two different batches of larvae each. Experiments with the first batch started on day 8 dpf and ended on day 24 dpf. The second batch was tested starting on day 5 dpf and ending on day 21 dpf. Within these periods, at least four experiments were conducted each day for each group size (N = 1,2 and 4) and each strain (WT and AB). Each experiment was conducted as follows: The watch glass serving as experimental arena was taken out of the setup and cleaned. After putting it back in place it was filled with a small volume of system water (the final volume would later be adjusted, see below). The underside of the watch glass was submerged in water, both in order to keep the temperature constant during the experiments and to reduce reflections which can occur at the air-glass boundary due to the different refractive indices of glass and water. Zebrafish larvae of both sexes were taken out of the holding tank separately and placed in the arena. The sizes of the larvae were matched by eye. After all the larvae

were in place, the body length of each larva was determined using a custom made Matlab script. First, the script acquired a camera frame. In this frame, we manually selected the the snout, the caudal peduncle and the tail of all the larvae in order to calculate their body length (Parichy et al. [29]). The frame was skipped if one or more larvae did not adopt a straight posture. In continuation, another frame was acquired and the procedure was repeated until a total number of four measurements had been made (i.e., only one frame was used for N=4, two for N=2, and four for N=1). From these measurements, the mean body length of the members of the group was calculated. In the next step, the volume of the water in the arena was adjusted. Due to the slope of the sides of the watch glass, a change in water volume and thus water depth also changed the surface area and the diameter of the arena. This was used to keep the diameter of the arena at approximately 20 BL by increasing the water volume as larvae grew larger. From the previously obtained mean body length, the corresponding diameter of the arena was calculated, and a circle of corresponding radius was superimposed on the live camera image in order to mark the desired borders of the arena. System water was then added or removed until the goal was met. To take advantage of the full resolution of the camera, the height of the camera, which was suspended directly above the arena, was adjusted until the arena covered the whole image. Together with the fact that the arena dimensions were linked to the absolute body length, larvae have a fixed body length of approximately 50 pixels in the video frames. In order to be able to deduce the absolute body length rather than the (constant) value in pixels, a millimeter scale was positioned next to the arena and the pixels-to-centimeter ratio was calculated. The scale was removed before the experiments started.

We then started the recording of a 20 minute video for each trial. Trials were done during the whole day, alternating randomly between trials of different group sizes. After a larva had been used in a trial, it was placed in a separate holding tank for the rest of the day in order to avoid repetitions with the same animals on the same day. Larvae were reunited in the evening after the last trial of the day had finished.

2.4.3 Experimental Setup

The experimental setup consisted in a big external tank of dimensions 95x135x30 cm, containing 180 liters of system water at 27.5°C, filtered and heated by an external filter system (Fluval 406, Teco TR 5). This external tank contained two 12x12x30 cm holding tanks, submerged up to 2/3 of their height in the water of the external tank in order to keep the

temperature constant. A weak flow of fresh water was delivered directly from the external tank. The holding tanks were placed directly inside the experimental setup in order to avoid stressful changes of water and temperature when transferring the larvae to the experimental arena, and minimize the time needed for the transfer. An opaque sheet of Plexiglass between the holding tanks and the experimental arena guaranteed that larvae in the holding tanks would not get disturbed by the experiments.

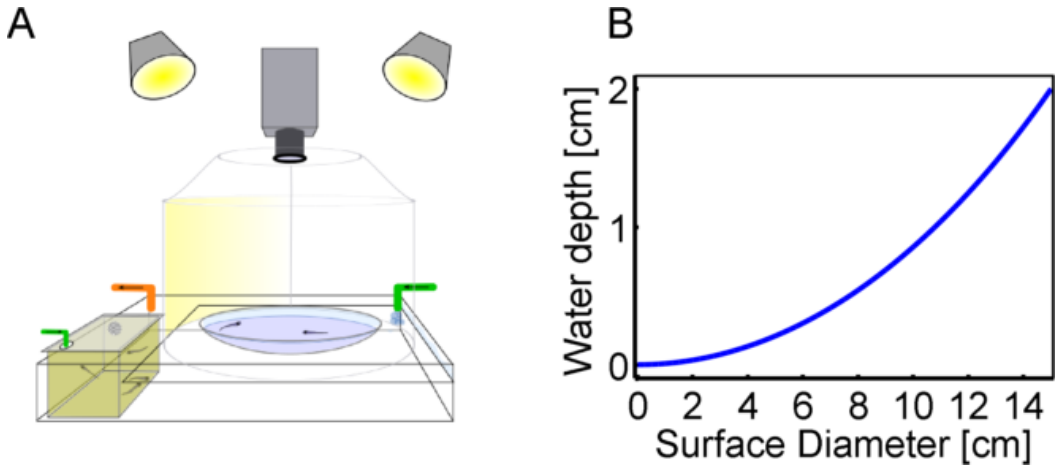

Fig. 2.11 *Water Depth Versus Surface Diameter* A) Sketch of the experimental setup B) Dependence of the water depth in the arena on the surface diameter.

The watch glass serving as experimental arena was placed at the center of a transparent Plexiglass board which was submerged beyond the water surface just enough to cover the the watch glass from below.
Videos were recorded by a Basler 622f camera with a Zeiss 16 mm objective, suspended directly above the watch glass, pointing downwards. The distance between the camera and the arena could be adjusted.
Before the start of each trial, a light diffusion dome was positioned over the arena to provide homogeneous light and avoid reflections and unwanted external stimuli, leaving only an opening for the camera on top.

2.4.4 Tracking and Trajectory Smoothing

The extraction of the coordinates of each individual in each video frame was done automatically by *idTracker*. Those frames in which the probability of the identity assigned to

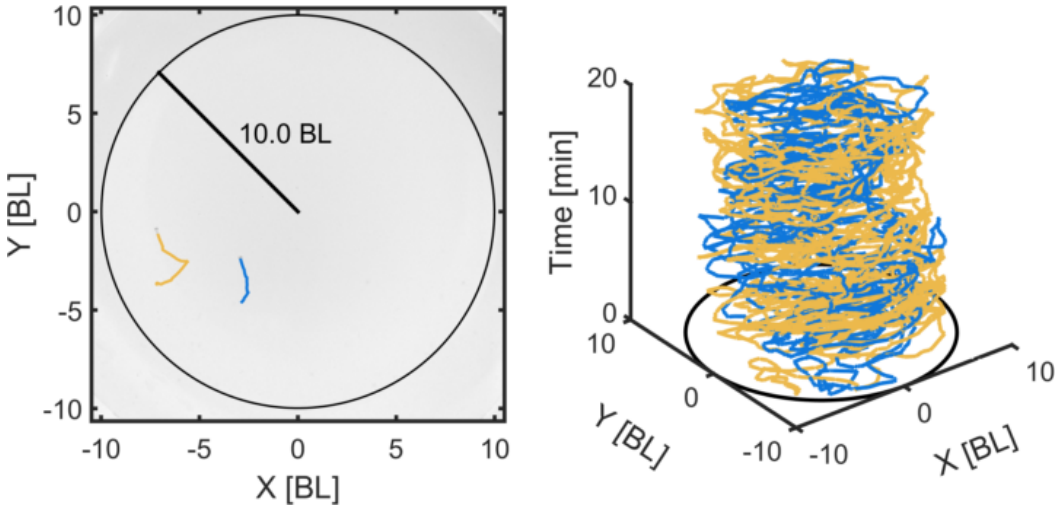

Fig. 2.12 *Example Frame and Tracking Result* A) Example frame for of an recording of two 20 dpf zebrafish. 2.5s long traces (blue/yellow) give an example of the tracking result. B) The full 20 min trajectories for each animal as obtained by *idTracker*.

an individual by *idTracker* was below 0.9 were neglected in all following analysis of this individual.

The position of each animal is calculated from the set of pixels representing the animal in a video frame. In order to reduce the positional information of an individual to one single x-y-coordinate, *idTracker* calculates the coordinate of each individual as the center of mass of all the pixels of the set corresponding to the animal. Since membership to the set of pixels is subject to noise due to spatial and temporal inhomogeneities in the illumination of the setup and fluctuations in the camera sensor, so is the center-of-mass coordinate.

Therefore, a careful assessment of the trajectory data is necessary. An additional difficulty arises from using the obtained trajectories not only in order to extract the position, but also the velocity and acceleration of each individual. These quantities are obtained from the positional data in form of the first and second time derivatives, and thereby inherit and amplify the noise.

In addition, changes in the body posture of an animal influence the position of the obtained coordinate with respect to the body of the animal. In the case of young zebrafish, the position of an animal usually coincides with the location of its head. Nevertheless, when the animal performs a sharp c-turn, the center of mass of all the corresponding pixels can even lie outside the contours of the body. Even less extreme movements like the undulation of an animal swimming forward can be imprinted on the extracted trajectory.

In our case, we are interested in acceleration behavior of zebrafish (acceleration and deceleration along the body axis and turning towards the right or left hand side) on a scale greater than the undulating frequency of the fish, i.e., we want to measure the overall course of an animal and not the more rapid body movements it needs to keep moving. Since the scale of noise in the measurement is even smaller (compare Figure B.3 with Figure B.5, especially first column), it will automatically be dealt with when smoothing applying.

A great number of smoothing methods for experimental data are available which are addressing this problem. In general, they use coordinates not only from a single frame, but use information from a number of frames before and/or after the frame in question in order to improve accuracy and smooth trajectories. We applied two of the most common methods in order to guarantee that our final results are not depending on the smoothing algorithm: A simple *Moving Average Smoothing* and *Cubic Spline Smoothing*. See SectionB.3 for details.

2.4.5 Randomized Controls

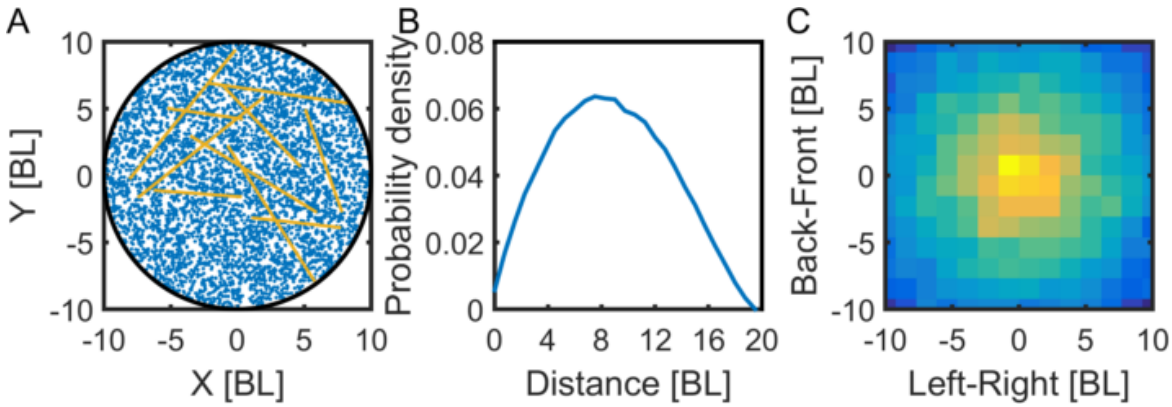

Fig. 2.13 *Distance Distribution and Relative Positions from Simulation* A Schematic example of the simulation. B) Distance distribution from pairs of randomly chosen coordinates. C) Probability of finding a conspecific at a position in the focal system obtained from simulation.

In order to determine the statistical significance of our results we need reliable controls against which we can compare our results. Since the majority of our analysis deal with social behavior, this means that we need control data by which we can asses if a certain behavior is socially motivated or not. This is not a trivial task. On the one hand there are many situations which can disguise non-social behavior as social behavior. On the other hand however, individuals might behave differently in the presence of conspecifics than they would behave being on their own, even though their change in behavior cannot be classified

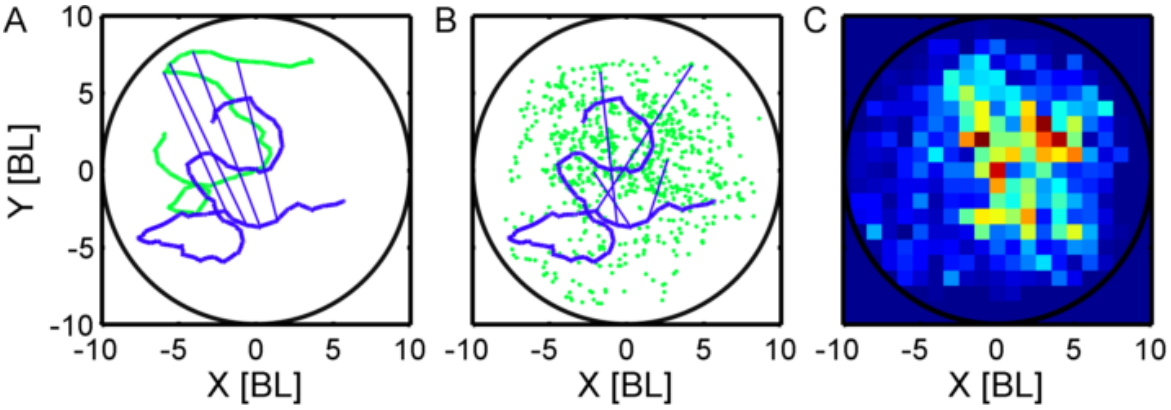

Fig. 2.14 Randomized Controls

as social. Results which at a first glance seem to confirm social interactions can be flawed by the geometry of the arena, as in the following examples:

- The borders of the arena reduce the freedom of movement of an individual and force it to turn away from the border and towards the center and thus towards other conspecifics present in the arena.

- There are gradients of light or food present in the arena which induce the individuals to prefer a certain area of the arena, or individuals prefer to stick to the walls of the arena. By this mechanisms group might form, but with our methods we cannot determine if this behavior has a social component or not.

Non-social changes in behavior can occur when individuals increase their activity in the presence of conspecifics, which, even though it could be attributed to social stimuli, might also be due to the general increase of stimulation in the arena. The latter is important because it makes it more difficult to use data from lone individuals as controls for data from groups. We therefore developed a scheme in which we obtain control data by randomizing the original data. For the majority of the analyses we proceeded as follows:
We start with the smoothed trajectories of the N individuals of the group. For each of the N individuals of a group ('focal'), we take the (N-1) - trajectories of its conspecifics ('neighbors'). Then, while we leave the trajectory of the focal untouched, we randomly change the temporal order of the trajectories of its neighbors: While the original trajectories are formed by x-y-coordinates sorted following the order of the video they are extracted from, i.e., starting with frame 1 and monotonically increasing to the last frame, x-y-coordinates for the randomized trajectories of the neighbors have a random order. Thus, for any given

frame, the x-y-coordinate of the focal for a given frame t_1 will coincide with the coordinate of a neighbor which in general originally corresponded to a different, earlier or later frame t_2. In order to omit any remaining correlation between the focal trajectory and its neighbors, we make sure that, originally, t_2 and t_1 were separated by at least 12000 frames, corresponding to about 6.6*min* in our experiments.
Note that we cannot calculate any meaningful velocities (and therefore accelerations) for the randomized neighbors, because their x-y-coordinated jump randomly between frames, and velocities are calculated from the difference vectors of the coordinates in two adjacent frames. For the majority of our analyses however, we only consider the position of the neighbor. As the trajectory of the focal is not changed, we can consider the velocity, acceleration or any quantity derived from it and study it in relation to the position of the neighbors.
For most types of analyses, this method also allows to repeat calculations with more randomized neighbors than present in the original experiment, and thus to generate more reliable control data.
A different method was used only for the analysis of the duration of pairwise interaction. Here, a x-y-coordinate was randomly chosen from the real trajectory of the focal. Then, occurrences of the neighbor entering a radius of 6 BL from the chosen coordinate at a later or earlier moment not corresponding to the real interaction (i.e., when the focal was not anymore or not yet present at the given coordinate) were detected and the duration of the uninterrupted stay within a distance of 6 BL from the virtual focal was calculated.

2.4.6 Calculation of Score

For individuals, the score of social behavior shown in Figure 2.9 for each type of analysis (inter-individual distance, social turning and attraction) is calculated from the p-value as

$$Score = 1 - \min(\alpha, p)/\alpha \tag{2.1}$$

where the significance level is $\alpha = 0.05$.
The p-value is obtained by comparing the mode of the distribution of inter-individual distance, the ratio of turns towards the conspecific or the ratio of attraction with controls from randomization, using a Monte-Carlo test with 10000 repetitions. The combined score is

given by the mean score

$$Score_{total} = (Score_{Distance} + Score_{turning} + Score_{attraction})/3. \tag{2.2}$$

Chapter 3

General Discussion

Our results show that zebrafish as a model organism for social behavior has the capacity to bring new insight into a number of aspects of social behavior in vertebrates. Especially the development of social behavior in zebrafish larvae offers the possibility of treating questions about the physiological and neurological requirements necessary for social behavior and the implications of a varying degree of interaction strength on collective behavior. In the course of this thesis, we have treated social behavior in zebrafish from both an experimental and a theoretical perspective. On the experimental side, we have introduced a new experimental paradigm for the measurement of freely behaving zebrafish larvae, we have used state-of-the-art techniques for data acquisition and have applied a series of tools to quantify social interactions. Finally, we found a basic mathematical model which describes social behavior in groups of young zebrafish and helps to interpret the mechanisms which lead to shoaling behavior.

Social behavior in zebrafish has been studied before using experimental arenas in which social behavior was detected by a higher probability of an animal being close to a transparent wall behind which it could perceive a group of conspecifics (Dreosti et al. [12], Hinz et al. [17]). This method has the advantage of offering an easy way to measure the strength of social behavior on a population level by a single parameter. However, excluding all social cues except visual stimuli, and limiting the movement of animals involved in social interactions, these kind of setups are not adequate for the study of more complex social interactions present in shoaling. Thus, in order to study the development of social behavior and a possible transition to more complex collective interactions in a group of zebrafish, we introduced a novel experimental arena, in which animals can move around and interact freely. Data acquisition and the characterization of social behavior in such a setup is more

complicated than in the above experimental paradigm, but has been made possible by great advances that have been made in recent years towards an automation of data acquisition and analysis of animal behavior. Concerning the behavior of free-swimming fish, *idTracker*, a software developed in our laboratory, has previously been used for the extraction of the coordinates of freely moving adult zebrafish from video recordings (Pérez-Escudero et al. [30]). In this thesis, we have shown that, with the right experimental conditions, trajectories can be extracted even for very young zebrafish larvae.
Trajectories of animals moving freely in a group store a large amount of information about social interactions, and methods are needed which are able to extract this information. This does not only apply to shoaling behavior only, but for all sorts of zebrafish behavior like aggression and dominant behavior. Only a small subset of the amount of behavioral traits (see Kalueff et al. [19]) is covered by the methods presented in this thesis, but many more can be defined and quantified on the basis of trajectories extracted from recordings, and the quantities which can be derived from them. One virtue of the toolbox *idSocial* presented in Chapter 1 is the easy integration of new types of analysis.

The development of zebrafish is not complete after three weeks, and also social behavior develops further (Buske and Gerlai [9]). The social interactions observed at early stages seem very different from those found for older animals. While young larvae move little and spend much time on what seems to be a careful inspection of their environment and their conspecifics, older animals show continuous movements and coordinate their movement, which requires instantaneous recognition of their conspecifics. But what happens between these two extremes in the development of social behavior? What does the change in the behavior of individuals mean for the behavior and movement as a group? Is there a sharp transition, with a clear change in the rules which govern social interactions, for example in the way individual count and take into account their conspecifics?

An indicator for a fundamental change in social interactions could for example be a transition from merely pairwise interactions at early stages of development to more complicated interactions involving three or more animals at once. We find only a weak sign for such a transition towards collective behavior, observable as an abrupt increase of the probability of a group size of four animal (i.e., including all individuals present in the experiment) (Figure 2.8F) at an age of 21 dpf.

There is a multitude of possible mechanisms leading from social interactions to collective behavior: It might be a gradual development in which the rules of interaction do not change,

but interactions get stronger. This might involve quantities and mechanisms which have not been considered in this thesis, for example the magnitude of acceleration towards a conspecific. This gradual increase in social behavior could still lead to a "phase transition" between lose social interactions and more coordinated group behavior for, example when interaction strength reaches a critical value above of which the nature of group behavior changes.A different mechanism leading to the robust shoaling and schooling behavior might depend on additional physiological changes which occur after the first social interactions can be observed. These changes might be similar to the one observed by Dreosti et al. [12] and in this work, concerning the preferred relative position of the neighbor, or more precise, its position in the visual field of the focal individual, which might have a direct impact on the coordination of movements and group movements (Perna et al. [31]) Finally, robust collective behavior can be a combination of both gradually increasing interaction strength, as we find for example for the probability of an individual turning towards a neighbor (Figure 2.5), for which we do not observe any abrupt change in behavior with age, and additional fundamental physiological changes.

One way to answer questions about the cognitive aspects of collective behavior comes from the study of the neural circuitry connected to social behavior. Since we find social interactions between animals at an age of 8 dpf, and individuals behaving "more social than random" even at 6 dpf, neuroimaging and opto-genetic manipulation can be used in order to study the functionality of brain regions or groups of neuron in social interactions. On the other hand, by manipulating individual behavior, we can get new insight into the interplay between individual and collective behavior.

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Appendix A

idSocial: Examples

A.1 Structure of *idSocial*

The main idea behind *idSocial* is to provide a set of Matlab® functions which can easily be applied to the whole set of data to be analyzed.

The available methods of analysis can be divided into the areas
Kinematics (Basic, non-social measures of movement like speed, acceleration etc. of an individual),
Interaction (Basic measures of social behavior like inter-individual distance),
Dynamics (Methods which analyze individual behavior connected with social interaction),
Hierarchies (Analysis of aggression, Territoriality and leader-follower relationship), and
Collective (Advanced analysis of group behavior).

Each *idSocial*-method takes the same three input parameters (given in the format of Matlab® structures): The first parameter contains the location of the trajectory data, along with some default options and additional optional information like the frame rate of each video. Each *idSocial* function will return the same structure, updated by the new results. The second parameter contains specific options for the current analysis, and the third determines how to reorder and/or pool the results, and the kind of statistics which should be applied.

In addition, there exist a number of auxiliary functions which can have different input parameters. The most important is *idSocial_loadData*, which takes a list containing the location to all of the trajectory data, a second (optional) list with additional information for each experiment, and options concerning the pre-processing of the data.

A typical *idSocial* analysis involves three major steps. First, the data set of pre-processed trajectories is obtained via *idSocial_loadData*. In the second step, the specific method of analysis is called for the data set. And last, the results obtained for the set of trajectories are re-ordered by combining the results from various trials and statistical quantities like the mean, the median etc are calculated.
The results can then be inspected and displayed with standard Matlab® functions or using additional *idSocial* routines.

Appendix B

Emergence of Social Behavior in Zebrafish Larvae, Additional Information

B.1 List of Experiments and Tracking results

The column titled *Tracking Status* contains measures of the quality of the trajectories obtained by *idTracker*. For each tracked frame and individual, *idTracker* returns the reliability that the identity as been assigned correctly, given by a value between 0 and 1. The numbers given in the table refer to mean value of the reliability, multiplied by 100 (*"Mean"*), the percentage of frames for which the reliability is 1 for all individuals (*"100"*), and the percentage of frames for which the reliability is at least 0.95 for all individuals (*"95"*).

Strain	No of fish	Start of exp.	Age (dpf)	Trial	Standard length (pxl)	Full body length (pxl)	Pixel per cm	Arena radius (pxl)	Dish size (cm)	Duration (min)	TrackingStatus (Mean/100/95)	Body length (idTracker)	Body width (idTracker)	Body area (idTracker)	Full body length (mm)
WT20141103	1	11.11.2014 17:54	8	1	36,8	22	21,1	388,62	15	00:20:15	100/100/100%	28,1	7,1	112	10,43
WT20141103	4	11.11.2014 18:35	8	1	39	22	23	447,51	15	00:20:06	100/21/96%	25,9	7	102,8	9,57
WT20141103	2	11.11.2014 19:19	8	1	40,3	22	20,5	414,23	15	00:20:06	100/56/100%	23,2	7,3	95,8	10,73
WT20141103	2	12.11.2014 17:58	9	2	44,3	51,8	126,8	496,47	15	00:20:15	100/52/86%	23,4	7,3	96,1	4,09
WT20141103	4	12.11.2014 18:47	9	2	44,3	51,5	124,1	487,21	15	00:20:07	95/0/18%	23,2	7,3	96,3	4,15
WT20141103	2	13.11.2014 12:56	10	3	43,5	52,1	117,5	439,77	15	00:20:07	100/41/92%	19,5	8,1	82,2	4,43
WT20141103	1	13.11.2014 13:31	10	2	40,4	46,9	115,7	420,6	15	00:20:07	100/100/100%	23,3	6,2	81,3	4,05
WT20141103	4	13.11.2014 14:06	10	3	42	49,7	117,4	434,11	15	00:20:08	99/7/84%	25	6,9	94,4	4,23
WT20141103	2	13.11.2014 15:41	10	4	39,7	47,7	118,6	457,29	15	00:20:06	100/25/96%	25,3	6,2	89,9	4,02
WT20141103	4	13.11.2014 16:17	10	4	42	49,2	114,2	467,44	15	00:20:06	92/4/43%	24,8	7	101,8	4,31
WT20141103	1	13.11.2014 16:48	10	3	44	51,6	117,4	483,43	15	00:20:06	100/100/100%	27,5	6,6	103,1	4,40
WT20141103	2	14.11.2014 12:03	11	5	43,4	50	112,6	478,56	15	00:20:05	97/14/57%	23,6	6,6	83,6	4,44
WT20141103	4	14.11.2014 12:39	11	5	43,3	50,2	114,1	478,37	15	00:20:06	82/0/13%	20,5	6,5	76,8	4,40
WT20141103	1	14.11.2014 13:12	11	4	44	51,7	114,6	436,7	15	00:20:07	100/87/87%	23,4	6,8	86,5	4,51
WT20141103	4	14.11.2014 13:43	11	6	42	48,6	112,6	438,78	15	00:20:05	98/1/35%	23,2	6,6	80	4,32
WT20141103	2	14.11.2014 16:24	11	6	44,2	50,8	107,2	461,35	15	00:20:22	97/6/69%	22	6,8	77,7	4,74
WT20141103	1	14.11.2014 17:05	11	5	41,7	49,7	112,5	460,12	15	00:20:07	100/83/83%	23,5	6,9	89,7	4,42
WT20141103	1	14.11.2014 17:54	11	6	42,9	50,4	116	470,38	15	00:20:05	100/79/79%	16	6,6	63	4,34
WT20141103	4	14.11.2014 18:30	11	7	46,1	53,3	111	489,12	15	00:20:04	90/0/16%	22,7	6,7	81,3	4,80
WT20141103	2	14.11.2014 19:53	11	7	41,3	48,7	114,9	478,64	15	00:20:04	100/20/92%	23,4	6,5	89,9	4,24
WT20141103	4	15.11.2014 14:13	12	8	44,7	52,8	114,6	485,07	15	00:20:06	92/1/48%	23,2	7,7	100,8	4,61
WT20141103	1	15.11.2014 14:48	12	7	44,8	52,8	107,2	492,32	15	00:20:04	100/100/100%	23,4	7,4	100,6	4,93
WT20141103	2	15.11.2014 15:17	12	8	49	56,9	107,4	489,59	15	00:20:05	76/0/26%	22,6	7,1	103,1	5,30
WT20141103	4	15.11.2014 15:46	12	9	45,2	53,5	106,9	467,07	15	00:20:06	98/29/72%	29,5	8,1	149,4	5,00
WT20141103	2	15.11.2014 16:21	12	9	45,6	52,1	104,1	469,22	15	00:20:05	99/27/66%	22	7,4	98,5	5,00
WT20141103	1	15.11.2014 16:52	12	8	43,1	50,3	108,9	490,64	15	00:20:05	100/99/99%	26,3	7,4	114,2	4,62
WT20141103	1	17.11.2014 11:54	14	9	49,9	56,6	102	467,94	15	00:20:05	100/99/99%	28,4	7,4	117,4	5,55
WT20141103	4	17.11.2014 12:31	14	10	46,5	52	103,9	474,68	15	00:20:04	57/0/1%	20,7	6,9	80,4	5,00
WT20141103	2	17.11.2014 13:01	14	10	44,5	50,9	101,2	491,94	15	00:20:05	95/0/48%	21,8	6	73,6	5,03
WT20141103	1	17.11.2014 13:36	14	10	47,5	51,8	100,3	465,1	15	00:20:05	100/98/98%	25,2	6,3	88,1	5,16
WT20141103	2	17.11.2014 14:44	14	11	45,8	50,5	109,2	449,72	15	00:20:05	100/90/98%	34,2	8,5	163,7	4,62
WT20141103	4	17.11.2014 15:27	14	11	45,8	50,8	98,5	454,15	15	00:20:05	97/18/70%	33,2	8,2	156,2	5,16

WT20141103	1	17.11.2014 15:53	14	11	45,9	51,8	100,5	463,27	15	00:20:04	100/100/100%	26,9	8,2	127,8	5,15
WT20141103	2	17.11.2014 16:22	14	12	41,5	47,3	102,2	461,03	15	00:20:05	98/39/80%	27,7	7,5	119,6	4,63
WT20141103	1	17.11.2014 17:37	14	12	46,6	53,7	98	461,02	15	00:20:04	100/99/99%	31,1	8,2	142,6	5,48
WT20141103	4	17.11.2014 18:13	14	12	45,2	53,1	100,4	466,27	15	00:20:04	100/18/98%	29,4	7,6	129,8	5,29
WT20141103	4	17.11.2014 18:46	14	13	43,5	49,7	98,5	478,13	15	00:20:05	99/28/89%	29,8	8,2	143,2	5,05
WT20141103	2	18.11.2014 11:03	15	14	44,9	50,2	98,6	476,1	15	00:20:06	100/75/95%	26,2	7,6	118,4	5,09
WT20141103	4	18.11.2014 11:41	15	14	43,2	48,7	95,1	443,58	15	00:20:05	100/33/89%	26,2	7,3	110,1	5,12
WT20141103	1	18.11.2014 12:08	15	13	47,8	54	98,6	466,26	15	00:20:05	100/100/100%	29,4	8,4	143,1	5,48
WT20141103	2	18.11.2014 12:47	15	15	42,6	50,1	86,4	456,73	15	00:20:05	100/97/100%	29	8,2	139,8	5,80
WT20141103	1	18.11.2014 13:18	15	14	44,5	51,6	86,9	473,61	15	00:20:05	100/99/99%	26	8,1	128,6	5,94
WT20141103	4	18.11.2014 13:54	15	15	41,6	48,4	92,9	449,75	15	00:20:05	100/58/99%	28,7	7,4	121,8	5,21
WT20141103	1	18.11.2014 14:25	15	15	34,4	40,3	93,4	388,62	15	00:20:05	100/100/100%	27,2	6,4	99	4,31
WT20141103	2	18.11.2014 15:04	15	16	39,4	45,3	91,7	445,26	15	00:20:12	100/95/99%	29,2	7,3	118,7	4,94
WT20141103	1	18.11.2014 15:36	15	16	34,8	40,9	93,7	405,94	15	00:20:05	100/100/100%	30,4	6,9	113,7	4,36
WT20141103	2	18.11.2014 17:45	15	17	43,6	51,9	93,6	455,36	15	00:20:05	100/94/99%	32,8	7,8	151,5	5,54
WT20141103	4	18.11.2014 18:14	15	16	37,9	43,8	93,7	417,19	15	00:20:03	100/9/96%	24,1	7,1	93,3	4,67
WT20141103	4	18.11.2014 18:52	15	17	38	45,7	93,5	445,96	15	00:20:05	100/9/83%	26,7	7	103,2	4,89
WT20141103	4	19.11.2014 11:05	16	18	42,5	49,3	93,4	491,16	15	00:20:06	100/10/89%	26,7	8,1	122,1	5,28
WT20141103	2	19.11.2014 11:37	16	18	38,5	45,5	91,9	429,88	15	00:20:05	100/53/100%	26,2	8	120,2	4,95
WT20141103	1	19.11.2014 12:18	16	17	35,6	41,2	92	408,67	15	00:20:04	100/100/100%	27	7	104,4	4,48
WT20141103	2	19.11.2014 12:44	16	19	39,8	48,4	93,3	447,64	15	00:20:05	100/35/87%	23,9	8	109,5	5,19
WT20141103	4	19.11.2014 13:14	16	19	41,7	49	93,5	436,45	15	00:20:07	99/19/93%	28,2	7,8	123,1	5,24
WT20141103	1	19.11.2014 13:39	16	18	43	49,7	93,4	468,83	15	00:20:05	100/100/100%	29	9,2	135,6	5,32
WT20141103	2	19.11.2014 14:04	16	20	41,8	50,5	93,4	452,98	15	00:20:05	100/53/97%	27,3	8,5	127,5	5,41
WT20141103	4	19.11.2014 16:29	16	20	42,5	51,5	93,4	477,77	15	00:20:03	99/50/94%	33,3	8,3	156,1	5,51
WT20141103	1	19.11.2014 16:59	16	19	47	54,8	89,9	468,3	15	00:20:05	100/100/100%	33,1	8,4	159,3	6,10
WT20141103	4	19.11.2014 17:31	16	21	38	45,2	91,9	454,97	15	00:20:05	100/44/99%	26,9	7,7	111,5	4,92
WT20141103	2	19.11.2014 17:58	16	21	37,7	45,6	89,9	454,12	15	00:20:05	100/87/100%	27	6,9	101,7	5,07
WT20141103	1	19.11.2014 18:26	16	20	38,7	46,9	91,6	426,22	15	00:20:05	100/100/100%	23,2	7,6	104,8	5,12
WT20141103	1	20.11.2014 10:36	17	21	36,5	43,2	95,1	396,53	15	00:20:06	100/100/100%	22,2	7,8	101	4,54
WT20141103	4	20.11.2014 11:04	17	22	47,9	56	96,8	475,02	15	00:20:06	99/34/90%	31,5	8,7	152,4	5,79
WT20141103	2	20.11.2014 11:30	17	22	47,6	54	95,1	478,5	15	00:20:05	100/84/93%	30,7	8,2	147,1	5,68
WT20141103	1	20.11.2014 11:57	17	22	43,3	50,7	96,9	477,28	15	00:20:06	100/100/100%	30,4	8,1	145,3	5,23
WT20141103	4	20.11.2014 12:33	17	23	39,5	46	83	460,8	15	00:20:04	100/73/99%	27,4	8,2	135,3	5,54
WT20141103	2	20.11.2014 13:04	17	23	41,7	48,7	91,5	466,44	15	00:20:05	100/76/96%	28,6	7,5	119,5	5,32
WT20141103	1	20.11.2014 13:48	17	23	39,6	46,6	91,3	415,48	15	00:20:05	100/98/98%	22	7,3	97,5	5,10
WT20141103	4	20.11.2014 14:32	17	24	42,8	51	92,9	463,15	15	00:20:06	99/3/82%	25,2	7,7	105,5	5,49

WT20141103	2	20.11.2014 15:00	17	24	43,8	52,3	93,2	472,6	15	00:20:06	100/79/98%	26,7	8,3	121,6	5,61
WT20141103	1	20.11.2014 15:25	17	24	44,2	51,4	89,2	482,31	15	00:20:05	100/100/100%	25,7	8,1	112,7	5,76
WT20141103	2	20.11.2014 17:33	17	25	41,2	49,6	81,3	464,25	15	00:20:06	100/97/100%	28,6	7,5	125,7	6,10
WT20141103	4	20.11.2014 18:05	17	25	42,3	49	85,6	452,33	15	00:20:06	100/47/94%	30,3	7,7	134,9	5,72
WT20141103	1	20.11.2014 18:45	17	25	39,8	47,9	88,2	412,47	15	00:20:08	100/100/100%	23	7,4	93,8	5,43
WT20141103	1	21.11.2014 10:54	18	26	40,6	47	90,3	406,51	15	00:20:07	100/100/100%	22,5	7,6	96	5,20
WT20141103	4	21.11.2014 11:26	18	26	43,2	50,9	85,8	448,18	15	00:20:05	100/26/94%	27,8	8,3	136,2	5,93
WT20141103	2	21.11.2014 11:53	18	26	41,7	49,1	86,9	437,35	15	00:20:06	100/36/88%	27,2	8,1	124,1	5,65
WT20141103	2	21.11.2014 12:18	18	27	43,5	50,7	86,7	453,65	15	00:20:05	100/57/98%	26,9	8	125,1	5,85
WT20141103	1	21.11.2014 12:46	18	27	39,1	45,9	90,7	420,07	15	00:20:05	100/100/100%	25,2	7,4	106,2	5,06
WT20141103	4	21.11.2014 13:35	18	27	46,1	52,8	93,3	497,82	15	00:20:05	99/6/65%	27,1	7,9	120,3	5,66
WT20141103	2	21.11.2014 14:09	18	28	46,2	52,8	88,8	476,84	15	00:20:04	100/3/89%	27,3	8	117	5,95
WT20141103	4	21.11.2014 14:34	18	28	46,8	53,7	87,3	480,18	15	00:20:05	100/46/95%	30,5	7,9	139,7	6,15
WT20141103	1	21.11.2014 16:52	18	28	45,8	52,2	88,2	477,1	15	00:20:05	100/100/100%	37,3	7,6	178,4	5,92
WT20141103	1	21.11.2014 17:26	18	29	47,3	54,3	88,2	468,32	15	00:20:04	100/100/100%	25	7	114,2	6,16
WT20141103	4	21.11.2014 18:24	18	29	47,1	52,3	85	475,77	15	00:20:05	100/55/89%	34,1	8,2	181,5	6,15
WT20141103	2	21.11.2014 18:50	18	29	41,4	47,3	83	439,71	15	00:20:06	100/47/88%	24,9	7,6	113,1	5,70
WT20141103	4	22.11.2014 11:43	19	30	46,8	53,9	83,4	478,83	15	00:20:06	100/72/100%	32,5	8,6	172,3	6,46
WT20141103	1	22.11.2014 12:15	19	30	43,9	49,2	83,1	462,81	15	00:20:05	100/100/100%	26,6	7,8	125,9	5,92
WT20141103	2	22.11.2014 12:48	19	30	39,8	46,4	88,2	469,8	15	00:20:06	75/0/7%	23,6	7,2	95,6	5,26
WT20141103	1	22.11.2014 13:14	19	31	45,9	52,6	84,7	473,99	15	00:20:05	100/100/100%	29,4	8,3	134,7	6,21
WT20141103	2	22.11.2014 13:43	19	31	44,2	49,8	81,7	451,76	15	00:20:05	100/84/98%	26,9	8,2	133,6	6,10
WT20141103	4	22.11.2014 14:13	19	31	40,1	46,5	83	455,96	15	00:20:05	93/2/51%	25,7	7,1	109,1	5,60
WT20141103	1	22.11.2014 15:05	19	32	43,3	50	72,6	466,02	15	00:20:20	100/100/100%	30,1	7,5	143,2	6,89
WT20141103	4	22.11.2014 15:37	19	32	42	48,8	82,4	449,36	15	00:20:05	99/10/84%	30,2	7,6	131,3	5,92
WT20141103	2	22.11.2014 16:09	19	32	39,9	46,7	85,1	468,79	15	00:20:05	100/33/96%	23,8	7,2	99,5	5,49
WT20141103	4	22.11.2014 16:41	19	33	41,8	48,3	85,8	471,56	15	00:20:05	99/4/82%	30,7	7,7	128,5	5,63
WT20141103	2	22.11.2014 17:43	19	33	42	49,1	90,5	455,32	15	00:20:05	100/42/94%	27,7	7,6	109,8	5,43
WT20141103	1	22.11.2014 18:20	19	33	43,3	50,7	79,5	453,65	15	00:20:05	100/100/100%	26,7	7,8	112,4	6,38
WT20141103	1	23.11.2014 11:32	20	34	40,2	44,8	84,7	441,32	15	00:20:06	100/100/100%	33,7	7,4	148,5	5,29
WT20141103	4	23.11.2014 11:59	20	34	43,5	49,5	86,4	466,8	15	00:20:05	98/0/55%	29	8,1	135,4	5,73
WT20141103	2	23.11.2014 12:34	20	34	43,6	49,5	88,3	489,57	15	00:20:05	99/72/90%	30,2	8,2	142,7	5,61
WT20141103	1	23.11.2014 13:03	20	35	40,4	45,9	86,4	457,57	15	00:20:04	100/99/99%	30,7	8	145,9	5,31
WT20141103	2	23.11.2014 13:30	20	35	39,8	46,1	86,1	460,03	15	00:20:06	99/42/91%	25,5	7,7	114,1	5,35
WT20141103	4	23.11.2014 14:11	20	35	46,8	55,1	79,6	464,46	15	00:20:05	100/52/97%	32,4	7,9	159,9	6,92
WT20141103	1	23.11.2014 14:49	20	36	45	50,6	70,9	453,07	15	00:20:01	100/100/100%	31	7,9	149,9	7,14
WT20141103	2	23.11.2014 15:17	20	36	39,8	46,3	86,5	460,02	15	00:20:05	100/64/100%	30,6	8,2	154,1	5,35

WT20141103	4	23.11.2014 15:53	20	36	43,1	50,6	77,3	468,84	15	00:20:05	100/75/99%	28,6	7,6	127,4	6,55
WT20141103	4	23.11.2014 16:30	20	37	46,7	51,2	77,8	454,01	15	00:20:04	100/93/100%	30,2	8,3	149,2	6,58
WT20141103	2	23.11.2014 17:00	20	37	42,1	48,2	88,2	466,24	15	00:20:06	100/93/98%	25,7	8	114	5,46
WT20141103	1	23.11.2014 17:30	20	37	49,5	55,6	86,4	478,44	15	00:20:04	100/100/100%	30,2	8,2	141,2	6,44
WT20141103	2	24.11.2014 11:08	21	38	43,6	48,5	84,8	467,09	15	00:20:06	100/87/100%	30,4	8,2	141,5	5,72
WT20141103	1	24.11.2014 11:37	21	38	44,8	50,2	86,4	460,99	15	00:20:05	100/100/100%	31,8	7,2	122,8	5,81
WT20141103	4	24.11.2014 12:06	21	38	44,3	50,5	84,9	474,54	15	00:20:05	99/46/95%	28	7,9	124,4	5,95
WT20141103	2	24.11.2014 12:40	21	39	49,6	54,9	69,3	474,3	15	00:20:06	100/66/98%	29,9	8,2	153,2	7,92
WT20141103	4	24.11.2014 13:14	21	39	46,2	51	86,6	472,54	15	00:20:05	100/57/97%	27,1	8,3	127,2	5,89
WT20141103	1	24.11.2014 15:15	21	39	40,7	44,5	72,6	413,55	15	00:20:04	100/100/100%	26,7	7,2	125,1	6,13
WT20141103	1	24.11.2014 16:17	21	40	48,2	52,6	72,8	445,5	15	00:20:06	100/100/100%	30,2	7,7	151,7	7,23
WT20141103	2	24.11.2014 16:45	21	40	42,6	47,5	72,8	442,83	15	00:20:06	100/100/100%	29,7	7,3	133,3	6,52
WT20141103	4	24.11.2014 17:17	21	40	45,6	50,1	81,3	472,05	15	00:20:05	100/41/98%	30,2	8,4	154,1	6,16
WT20141103	2	24.11.2014 17:45	21	41	48,7	53,7	72,6	479,51	15	00:20:05	100/100/100%	30,6	8,4	166,4	7,40
WT20141103	4	24.11.2014 18:18	21	41	45,6	51	74,5	469,07	15	00:20:05	100/83/98%	31,4	8,2	165,9	6,85
WT20141103	1	24.11.2014 18:46	21	41	48,6	54,2	72,8	484,03	15	00:20:05	100/98/98%	34,3	8,2	179,6	7,45
WT20141103	1	25.11.2014 10:41	22	42	48	53,6	72,8	473,72	15	00:20:05	100/100/100%	29,8	8,4	166,1	7,36
WT20141103	4	25.11.2014 11:12	22	42	42,4	46,5	79,7	464,74	15	00:20:07	100/85/100%	30,6	7,5	136,7	5,83
WT20141103	2	25.11.2014 11:49	22	42	43,2	48,1	83,1	441,61	15	00:20:31	100/92/100%	29,6	7,7	134,6	5,79
WT20141103	2	25.11.2014 12:21	22	43	43,5	49,7	74,5	467,58	15	00:20:06	100/100/100%	31	7,7	148,8	6,67
WT20141103	1	25.11.2014 12:50	22	43	40,9	45,5	78,3	441,52	15	00:20:05	100/100/100%	28,3	8	137,8	5,81
WT20141103	4	25.11.2014 13:16	22	43	42,7	47,9	78	447,43	15	00:20:06	100/36/95%	26,6	7,9	124,7	6,14
WT20141103	1	25.11.2014 14:02	22	44	46,1	51,5	76,2	446,25	15	00:20:05	100/100/100%	32,7	8	161,2	6,76
WT20141103	4	25.11.2014 15:00	22	44	47,3	52,1	71,1	486,39	15	00:20:05	100/95/100%	29,8	8,6	162	7,33
WT20141103	2	25.11.2014 15:31	22	44	45,1	51	80	468,67	15	00:20:04	100/100/100%	31,5	8,2	160,8	6,38
WT20141103	4	25.11.2014 17:19	22	45	42,5	46,9	66,8	442,56	15	00:20:04	100/97/100%	29	7,8	155,4	7,02
WT20141103	2	25.11.2014 17:51	22	45	47,2	51,5	67,5	470,16	15	00:20:05	100/100/100%	34	8,3	194,8	7,63
WT20141103	1	25.11.2014 18:19	22	45	48,5	53,9	64,5	463,87	15	00:20:05	100/100/100%	29,4	8,5	177,9	8,36
WT20141103	2	26.11.2014 10:55	23	46	51,5	56,8	65,8	483,54	15	00:20:07	100/100/100%	32,3	8,2	172,5	8,63
WT20141103	4	26.11.2014 11:26	23	46	47,8	53	63,1	485,19	15	00:20:04	100/95/99%	30,2	7,8	157,7	8,40
WT20141103	1	26.11.2014 12:04	23	46	42,9	46,8	63,1	438,96	15	00:20:05	100/100/100%	28,7	8,1	146,8	7,42
WT20141103	1	26.11.2014 12:39	23	47	38,8	43,3	66,8	378,57	15	00:20:05	100/100/100%	25,9	7,6	113,4	6,48
WT20141103	4	26.11.2014 13:07	23	47	42,9	46,7	64,3	434,26	15	00:20:05	90/33/54%	27,3	7,7	133,4	7,26
WT20141103	2	26.11.2014 13:36	23	47	46,2	50,1	67,5	466,88	15	00:20:05	100/95/97%	29,4	8	148,5	7,42
WT20141103	1	26.11.2014 14:03	23	48	38,9	43,5	65,7	423,34	15	00:20:05	100/100/100%	24,6	7,3	106,5	6,62
WT20141103	2	26.11.2014 15:24	23	48	42,3	48	81	461,17	15	00:20:05	100/97/100%	27,5	7,3	115,8	5,93
WT20141103	4	26.11.2014 16:31	23	48	47,4	51,6	79,8	490,61	15	00:20:04	100/92/99%	33	8,1	173,4	6,47

WT20141103	2	26.11.2014 17:05	23	49	46,9	52	67,8	463,54	15	00:20:05	100/100/100%	31	8	171,1	7,67
WT20141103	4	26.11.2014 18:01	23	49	44,8	48,8	69,2	447,06	15	00:20:06	100/89/99%	30,7	7,9	163,3	7,05
WT20141103	1	26.11.2014 18:28	23	49	45,6	50,4	68	459,8	15	00:20:05	100/100/100%	29,4	8,2	164,4	7,41
WTpetri20141103	1	27.11.2014 10:55	24	50	43,8	49,1	64,2	467,02	15	00:20:08	100/100/100%	32,7	8,5	166,3	7,65
WT20141103	1	27.11.2014 11:32	24	50	49,8	54,4	64	471,16	15	00:20:05	100/100/100%	30,1	8	156,1	8,50
WT20141103	2	27.11.2014 12:03	24	50	45,3	51	62,2	471,31	15	00:20:05	100/96/96%	27,4	7,5	135,5	8,20
WTpetri20141103	2	27.11.2014 12:38	24	50	44,2	49,8	64	476,17	15	00:20:04	100/99/100%	26,2	7,5	122,5	7,78
WTpetri20141103	4	27.11.2014 13:09	24	50	42,5	48,4	62,3	468,05	15	00:20:05	100/61/91%	26,7	7,9	131,3	7,77
WT20141103	4	27.11.2014 13:40	24	50	45,2	50,7	62,8	445,29	15	00:20:05	100/96/99%	26,4	7,5	127,7	8,07
WT20141103	4	27.11.2014 15:13	24	51	43,1	48,2	62,6	455,9	15	00:20:05	100/92/99%	27,8	7,3	137,8	7,70
WTpetri20141103	4	27.11.2014 16:24	24	51	40,8	44,9	64,2	453,69	15	00:20:05	98/46/82%	26,6	7,2	119,6	6,99
WTpetri20141103	1	27.11.2014 17:00	24	51	40,5	43,5	64	454,28	15	00:20:04	100/100/100%	25,8	7,1	114,4	6,80
WT20141103	1	27.11.2014 17:36	24	51	39	42	62,3	433,82	15	00:19:49	100/100/100%	27,4	6,9	119,8	6,74
WT20141103	2	27.11.2014 18:08	24	51	45,5	50,5	64	463,75	15	00:20:05	100/98/100%	28,5	7,7	149,2	7,89
WTpetri20141103	2	27.11.2014 18:34	24	51	39,8	43,8	62,6	459,65	15	00:20:04	100/98/100%	29,1	7,6	144,8	7,00
WT20141103	2	28.11.2014 11:29	25	52	50,7	56,1	64	460,71	15	00:20:05	100/98/99%	29,6	7,5	139,6	8,77
WTpetri20141103	2	28.11.2014 11:59	25	52	46,3	51,7	62,3	456,86	15	00:20:03	100/99/99%	26,1	7,4	115,5	8,30
WT20141103	1	28.11.2014 12:28	25	52	44,2	48,3	64	465,55	15	00:20:05	100/100/100%	24,6	6,8	106,6	7,55
WTpetri20141103	1	28.11.2014 12:54	25	52	38,5	43,7	62,3	454,96	15	00:20:08	100/100/100%	18,5	7	78,9	7,01
WTpetri20141103	4	28.11.2014 14:53	25	52	42,8	47,2	64,1	452,82	15	00:20:05	99/51/78%	32,8	8,1	172,1	7,36
WT20141103	4	28.11.2014 15:24	25	52	47,7	51,6	64	469,9	15	00:20:06	100/88/99%	31,9	8	171,6	8,06
WT20141126	1	01.12.2014 10:45	5	1	46,5	51,7	126,2	445,09	15	00:20:10	100/100/100%	49,1	6,5	178,3	4,10
WT20141126	2	01.12.2014 11:29	5	1	46	50,3	127,1	462,18	15	00:20:06	91/0/0%	40,7	6,6	148,3	3,96
WT20141126	4	01.12.2014 12:00	5	1	45,4	49,5	126,6	457,07	15	00:20:05	84/0/33%	36,2	6,7	126,4	3,91
WT20141126	2	01.12.2014 12:32	5	2	46,6	51,4	122,7	464,73	15	00:20:05	100/88/100%	38,1	6,8	134,5	4,19
WT20141126	1	01.12.2014 12:58	5	2	47,1	52,3	124,8	470,51	15	00:20:05	100/100/100%	44,2	6,6	144,5	4,19
WT20141126	4	01.12.2014 13:24	5	2	45,3	50,6	122,9	489,11	15	00:20:04	100/31/98%	38,2	6,5	127	4,12
WT20141126	1	01.12.2014 15:35	5	3	44,4	49,1	126,6	473,87	15	00:20:05	100/100/100%	37,9	6,9	133,2	3,88

WT20141126	2	01.12.2014 16:02	5	3	44,1	48,2	125,9	452,02	15	00:20:05	95/30/88%	38,8	6,8	132,6	3,83
WT20141126	4	01.12.2014 16:29	5	3	46,4	50,6	127,4	450,95	15	00:20:05	100/37/92%	36,9	6,9	123,5	3,97
WT20141126	2	01.12.2014 17:05	5	4	45,2	49,6	126,8	428,96	15	00:20:04	100/95/100%	35,6	6,7	123,6	3,91
WT20141126	1	01.12.2014 17:35	5	4	44,7	49,7	126,8	478,19	15	00:20:05	100/100/100%	31	6,9	113,1	3,92
WT20141126	4	01.12.2014 18:02	5	4	47,1	51,5	124,8	460,06	15	00:20:04	100/13/87%	31,8	6,9	113,9	4,13
WT20141126	1	02.12.2014 11:01	6	5	47,8	52,6	124,5	468	15	00:20:07	100/100/100%	35,8	6,9	124,5	4,22
WT20141126	2	02.12.2014 11:30	6	5	45,3	48,9	121,1	446,47	15	00:20:05	99/79/98%	33,9	6,8	119,2	4,04
WT20141126	4	02.12.2014 12:37	6	5	47,1	50,8	122,8	461,55	15	00:20:21	100/0/96%	34,4	6,7	118,5	4,14
WT20141126	2	02.12.2014 13:13	6	6	44,4	49,4	123,2	446,67	15	00:20:05	100/53/91%	27,3	6,8	99,7	4,01
WT20141126	4	02.12.2014 13:40	6	6	45,1	49,9	122,2	446,74	15	00:20:05	96/16/76%	34,8	6,7	116,1	4,08
WT20141126	1	02.12.2014 15:29	6	6	46,1	50,4	121,8	454,93	15	00:20:05	100/100/100%	36,6	6,6	120,8	4,14
WT20141126	4	02.12.2014 15:59	6	7	45,6	49,8	123,3	480,91	15	00:20:05	100/24/85%	29,8	7	106	4,04
WT20141126	1	02.12.2014 16:23	6	7	45,7	50,1	122,8	459,59	15	00:20:05	100/100/100%	30,1	6,8	105	4,08
WT20141126	2	02.12.2014 16:50	6	7	44,4	48,9	120,5	436,24	15	00:20:04	100/22/63%	35	6,5	117,4	4,06
WT20141126	2	02.12.2014 17:25	6	8	45,4	50,3	122,8	460,29	15	00:20:05	100/14/99%	32,9	6,8	108,7	4,10
WT20141126	4	02.12.2014 17:50	6	8	45,7	50,1	124,8	467,46	15	00:20:05	98/19/49%	29,2	7	104,8	4,01
WT20141126	1	02.12.2014 18:15	6	8	44,5	48,8	124,6	460,45	15	00:20:05	100/96/96%	26,2	7	99,7	3,92
WT20141126	4	03.12.2014 11:00	7	9	45,4	50,1	122,2	470,41	15	00:20:06	99/10/44%	32	7	112,5	4,10
WT20141126	1	03.12.2014 11:27	7	9	46,1	50,9	123,5	469,84	15	00:20:04	100/100/100%	31,5	6,8	115,4	4,12
WT20141126	2	03.12.2014 11:55	7	9	46,9	51,5	123,7	452,65	15	00:20:05	100/85/100%	31,7	6,9	115,7	4,16
WT20141126	1	03.12.2014 12:20	7	10	46,6	51,7	123,1	481,77	15	00:20:05	100/100/100%	26,9	7,2	105,3	4,20
WT20141126	2	03.12.2014 13:01	7	10	46,7	52	121	467,66	15	00:20:06	100/98/100%	28,7	6,9	104,9	4,30
WT20141126	4	03.12.2014 13:52	7	10	47,2	53	124,2	459,7	15	00:20:05	99/63/89%	27,9	7,2	104,2	4,27
WT20141126	4	03.12.2014 15:39	7	11	48	53	124,2	465,92	15	00:20:05	100/94/100%	30,4	7	110,8	4,27
WT20141126	2	03.12.2014 16:11	7	11	47,9	52,4	124,2	484,69	15	00:20:04	92/75/83%	32,3	7,2	117,6	4,22
WT20141126	2	03.12.2014 16:48	7	12	47,2	52	122,1	477,96	15	00:20:05	100/95/100%	27,3	7,1	110,4	4,26
WT20141126	1	03.12.2014 17:16	7	11	48,4	52,7	123,5	478,05	15	00:20:05	100/100/100%	32,6	7	132,3	4,27
WT20141126	4	03.12.2014 17:46	7	12	48,3	53,4	122,2	472,29	15	00:20:07	92/1/35%	30,5	7	120	4,37
WT20141126	1	03.12.2014 18:13	7	12	48,5	54,1	122,8	450,64	15	00:20:04	100/100/100%	28,1	7	111,2	4,41
WT20141126	2	04.12.2014 11:14	8	13	49	54,2	121,8	464,09	15	00:20:06	100/95/100%	26,3	7,4	109,4	4,45
WT20141126	1	04.12.2014 11:48	8	13	45,5	50,3	122	459,8	15	00:20:06	100/100/100%	25,3	7,3	106,1	4,12
WT20141126	4	04.12.2014 12:16	8	13	48,4	53,7	121,4	478,09	15	00:19:20	100/42/92%	29,9	7,4	121,6	4,42
WT20141126	1	04.12.2014 12:46	8	14	49,2	54,2	121,7	493,92	15	00:20:06	100/100/100%	24,1	7,4	104,1	4,45
WT20141126	4	04.12.2014 13:12	8	14	47,9	54,3	121	497,29	15	00:20:05	100/58/99%	29,7	7,4	122,7	4,49
WT20141126	2	04.12.2014 13:40	8	14	50,2	55,7	121,4	489,77	15	00:20:05	100/84/100%	27,5	7,5	117,5	4,59
WT20141126	4	04.12.2014 14:08	8	15	47,7	52,6	119,6	469,9	15	00:20:05	98/14/78%	25,8	7,5	106,9	4,40
WT20141126	2	04.12.2014 16:11	8	15	45,5	49,3	122,7	502	15	00:20:05	100/70/97%	22,5	7,5	92,9	4,02

WT20141126	1	04.12.2014 16:44	8	15	45,2	49,4	121,2	480,66	15	00:20:04	100/99/99%	23,3	6,8	88,1	4,08
WT20141126	2	04.12.2014 17:10	8	16	47,4	53,2	122,9	492,02	15	00:20:05	100/70/100%	23,2	7,4	98,1	4,33
WT20141126	4	04.12.2014 17:48	8	16	44,3	49,5	121	473,04	15	00:20:06	99/43/92%	23,7	7,4	95,8	4,09
WT20141126	1	04.12.2014 18:15	8	16	49,1	54,2	121	500,13	15	00:20:05	100/100/100%	21,8	8,1	96,6	4,48
WT20141126	4	05.12.2014 10:54	9	17	47,6	52,2	119,1	477,69	15	00:20:05	100/72/99%	26	7,3	109	4,38
WTpetri20141126	4	05.12.2014 11:33	9	1	40	44,7	103,7	461,09	8,7	00:20:05	99/75/90%	44,7	6,3	81,3	4,31
WT20141126	1	05.12.2014 12:03	9	17	49,3	53,6	117,7	493,82	15	00:20:05	100/100/100%	32,8	7,1	130,6	4,55
WT20141126	2	05.12.2014 12:40	9	17	47,1	52,5	118,8	481,51	15	00:20:05	100/95/100%	27,5	7,3	112,1	4,42
WT20141126	1	05.12.2014 13:05	9	18	49,1	54	117,7	465,59	15	00:20:04	100/100/100%	34,4	6,9	127	4,59
WT20141126	2	05.12.2014 14:04	9	18	46,2	51,1	117,4	472,07	15	00:20:06	100/97/100%	29,1	6,9	110,3	4,35
WT20141126	4	05.12.2014 15:37	9	18	47,6	52,5	117,6	473,13	15	00:20:04	100/59/97%	24,8	7,2	95,8	4,46
WTpetri20141126	4	05.12.2014 16:06	9	2	39,1	44,1	102,9	457,72	9	00:20:04	98/18/57%	19,3	6,5	68,2	4,29
WT20141126	2	05.12.2014 16:37	9	19	46,7	51,5	112,6	483,51	15	00:20:05	98/29/76%	21	7	83,7	4,57
WT20141126	4	05.12.2014 17:18	9	19	50,3	54,8	112,5	479,05	15	00:20:04	99/33/94%	25,9	7,5	109,6	4,87
WTpetri20141126	4	05.12.2014 17:54	9	3	42,4	47,7	104,4	464,53	9	00:20:04	99/58/79%	27,4	7,1	108,7	4,57
WT20141126	1	05.12.2014 18:21	9	19	50	54,5	112	486,1	15	00:20:05	100/99/99%	28,3	7,3	115,6	4,87
WT20141126	4	05.12.2014 18:51	9	20	46,5	52	111,9	465,86	15	00:20:05	100/50/87%	29,4	7,4	123,7	4,65
WTpetri20141126	4	05.12.2014 19:25	9	4	45	48,9	101,7	465,67	9	00:19:21	97/20/54%	28,5	7,1	110,8	4,81
WT20141126	2	05.12.2014 20:14	9	20	48,3	53,9	109,1	481,36	15	00:20:04	100/88/99%	29,8	7,4	125,9	4,94
WT20141126	1	05.12.2014 20:38	9	20	46,9	51,5	109,2	454,87	15	00:20:04	100/100/100%	28,9	7,4	125,2	4,72
WT20141126	4	06.12.2014 11:11	10	21	48	52,6	111,1	493,54	15	00:20:05	100/55/98%	31,7	7,8	143,1	4,73
WT20141126	1	06.12.2014 11:41	10	21	45,4	49,7	109,6	480,6	15	00:20:05	100/100/100%	32,2	7,2	129,8	4,53
WT20141126	2	06.12.2014 12:18	10	21	46,9	50	109	468,82	15	00:20:05	100/94/100%	33,7	7,6	150,9	4,59
WT20141126	2	06.12.2014 12:45	10	22	46,2	50,4	109,4	470,13	15	00:20:05	100/100/100%	27,4	6,9	112,8	4,61
WT20141126	2	06.12.2014 13:19	10	23	44	48,4	109,4	481,32	15	00:20:04	100/94/100%	23,1	7,2	97,7	4,42
WT20141126	1	06.12.2014 13:48	10	22	53,4	57,8	100	485,37	15	00:20:04	100/100/100%	33,5	7,7	143,2	5,78
WT20141126	1	06.12.2014 14:17	10	23	47,3	51	99	476,24	15	00:20:04	100/100/100%	31,4	7,4	142,4	5,15
WT20141126	4	06.12.2014 14:50	10	22	46,7	50,7	100,6	467,72	15	00:20:04	100/47/99%	30,2	7	126,9	5,04
WT20141126	4	06.12.2014 15:17	10	23	45	48,3	98,5	469,45	15	00:20:05	100/76/100%	29,2	7,1	123	4,90
WT20141126	2	06.12.2014 16:01	10	24	48,5	52,6	103,9	469,35	15	00:20:05	100/95/100%	30,4	7,4	136,4	5,06
WT20141126	4	06.12.2014 16:38	10	24	51,9	56,4	103,9	488,16	15	00:20:04	100/80/100%	31,3	7,7	143,8	5,43
WT20141126	1	06.12.2014 17:05	10	24	44,7	47,8	102,5	468,14	15	00:20:05	100/100/100%	29,1	7,1	118,8	4,66
WT20141126	1	07.12.2014 11:37	11	25	47,6	51,9	102,7	486,48	15	00:20:05	100/100/100%	26,2	7,7	113,1	5,05

WT20141126	4	07.12.2014 12:18	11	25	46,9	51,6	95,3	486,97	15	00:20:05	85/5/33%	29,9	7,1	119	5,41
WT20141126	2	07.12.2014 12:48	11	25	44,4	48,7	93,9	454,88	15	00:20:05	100/98/100%	28,8	6,8	111,7	5,19
WT20141126	2	07.12.2014 13:32	11	26	49,2	53,7	97,2	480,4	15	00:20:04	100/90/100%	27,2	7,4	122	5,52
WT20141126	1	07.12.2014 13:57	11	26	46,5	50,5	97	450,69	15	00:20:04	100/100/100%	22,3	7	90,1	5,21
WT20141126	4	07.12.2014 14:26	11	26	47,2	51,8	96,3	475,42	15	00:20:04	99/54/87%	26,3	7,4	114,3	5,38
WT20141126	1	07.12.2014 14:59	11	27	45,4	50,2	108,2	442	15	00:20:04	100/100/100%	24,3	7,2	98	4,64
WT20141126	4	07.12.2014 15:32	11	27	46,8	50,9	96,9	483,34	15	00:20:04	98/61/92%	29	7,2	126,8	5,25
WT20141126	2	07.12.2014 16:15	11	27	46,9	51,4	96,9	453,99	15	00:20:05	100/99/100%	27,1	6,8	112,5	5,30
WT20141126	4	07.12.2014 16:50	11	28	47	50,5	101,2	478,3	15	00:20:05	100/32/98%	28	7,1	118,1	4,99
WT20141126	2	07.12.2014 17:28	11	28	44,5	48,1	107,9	452,43	15	00:20:05	100/85/99%	26	6,4	99,1	4,46
WT20141126	1	07.12.2014 17:57	11	28	49,8	53,2	100,3	467,59	15	00:20:05	100/100/100%	25	6,9	103,2	5,30
WT20141126	4	07.12.2014 18:27	11	29	47,6	52,2	99,3	485,9	15	00:20:06	100/56/94%	31,3	7,3	133,4	5,26
WT20141126	1	08.12.2014 11:18	12	29	47,3	51,7	110,7	487,92	15	00:20:05	100/100/100%	23,1	7,8	98,8	4,67
WT20141126	4	08.12.2014 11:51	12	30	46,3	51,1	105,7	487,4	15	00:20:04	100/67/95%	25,8	7,4	109,7	4,83
WT20141126	2	08.12.2014 12:19	12	29	49,3	53,8	94,4	488,15	15	00:20:06	100/100/100%	29,7	7,3	124,2	5,70
WT20141126	1	08.12.2014 12:45	12	30	45,4	50,6	93,2	449,28	15	00:20:04	100/100/100%	28,8	7,1	118,2	5,43
WT20141126	2	08.12.2014 13:17	12	30	42,4	46,6	106	458,88	15	00:20:05	98/80/97%	24	6,6	91,4	4,40
WT20141126	4	08.12.2014 13:47	12	31	46,2	49,9	97	481,53	15	00:20:05	100/48/92%	24,1	7,2	102,6	5,14
WT20141126	2	08.12.2014 14:20	12	31	48,3	51,9	98,3	459,11	15	00:20:04	100/48/92%	51,9	7,4	109,6	5,28
WT20141126	4	08.12.2014 14:58	12	32	49	53,5	98	486,94	15	00:20:04	100/56/97%	32,7	7,5	143,6	5,46
WT20141126	1	08.12.2014 15:23	12	31	45,3	49,4	97,8	471,27	15	00:20:04	100/100/100%	27	6,9	113	5,05
WT20141126	4	08.12.2014 15:52	12	33	49,3	53,7	98,1	470,46	15	00:20:05	100/71/97%	30,8	7,5	134,2	5,47
WT20141126	2	08.12.2014 16:19	12	32	51	56,9	97,8	495,77	15	00:20:05	100/78/98%	31,4	7,6	134,7	5,82
WT20141126	1	08.12.2014 17:16	12	32	47,3	52,3	90,9	460,15	15	00:20:04	100/100/100%	23,1	7	89,3	5,75
WT20141126	4	08.12.2014 17:46	12	34	46,9	51,4	97	507,07	15	00:20:04	100/33/92%	30,7	7,4	131,6	5,30
WT20141126	12	08.12.2014 18:20	12	1	46,4	50,5	96	479,21	15	00:20:06	96/0/26%	25	6,9	95,5	5,26
WT20141126	1	09.12.2014 11:04	13	33	51,6	55,6	95,3	494,63	15	00:20:04	100/100/100%	34,8	8,5	166,1	5,83
WT20141126	2	09.12.2014 11:39	13	33	47,2	51,9	96,1	477,9	15	00:20:04	100/97/99%	28,1	7,4	117,6	5,40
WT20141126	4	09.12.2014 12:11	13	35	46,4	50,1	92,7	485,37	15	00:20:04	100/70/99%	28,6	7,6	122,6	5,40
WT20141126	4	09.12.2014 12:43	13	36	48,3	51,9	97	480,75	15	00:20:05	100/57/98%	29,1	7,5	123,1	5,35
WT20141126	1	09.12.2014 13:11	13	34	50,5	54,5	92,9	471,41	15	00:20:01	100/100/100%	24	7,2	95,1	5,87
WT20141126	2	09.12.2014 13:50	13	34	45,2	49,7	88,9	469,73	15	00:20:01	99/51/93%	27,2	7	104,6	5,59
WT20141126	1	09.12.2014 16:08	13	35	47,2	50,9	90,3	481,18	15	00:20:01	100/100/100%	32,5	6,9	123,3	5,64
WT20141126	2	09.12.2014 16:39	13	35	48,4	52,8	90,2	483,18	15	00:20:03	100/98/99%	29,3	7,7	138,2	5,85
WT20141126	4	09.12.2014 17:09	13	37	48,7	52,4	91,4	483,91	15	00:20:04	100/81/99%	29,3	7,3	129,5	5,73
WT20141126	2	09.12.2014 17:41	13	36	49,4	52,6	93,5	494,76	15	00:20:05	100/83/93%	28,9	7,2	118,1	5,63
WT20141126	4	09.12.2014 18:16	13	38	48,6	53,2	104,2	462,41	15	00:20:04	100/64/100%	30,3	7,6	127,5	5,11

WT20141126	4	09.12.2014 18:48	13	39	49,5	54	91	472,41	15	00:20:05	100/32/94%	28,2	7,4	122,4	5,93
WT20141126	1	09.12.2014 19:32	13	36	49,5	54	91	462,79	15	00:20:05	100/100/100%	23,3	6,9	91,1	5,93
WTcloseup20141126	2	09.12.2014 20:01	13	1	68,4	73,3	126,5	482,42	15	00:20:05	100/73/90%	46,1	9,8	266,4	5,79
WT20141126	4	10.12.2014 11:28	14	40	47,1	51,1	84,3	482,05	15	00:20:06	99/47/87%	51,1	7,4	121,9	6,06
WT20141126	1	10.12.2014 12:00	14	37	49,9	53,6	93,6	464,05	15	00:20:06	100/100/100%	53,6	8	102,6	5,73
WT20141126	2	10.12.2014 12:27	14	37	47,9	52	97,8	473,92	15	00:20:05	95/22/62%	52	7,7	97,9	5,32
WT20141126	2	10.12.2014 12:55	14	38	47,2	51,1	90,9	480,42	15	00:20:04	96/52/81%	51,1	7,6	107	5,62
WT20141126	4	10.12.2014 13:32	14	41	47,4	51	91,5	498,18	15	00:20:05	99/19/81%	51	8	117,4	5,57
WT20141126	1	10.12.2014 14:15	14	38	49,1	53,2	92,4	489,11	15	00:20:05	100/100/100%	23,7	8	102,6	5,76
WT20141126	1	10.12.2014 15:44	14	39	46,9	51	94,4	489,39	15	00:20:09	100/100/100%	22,2	7,8	97,8	5,40
WT20141126	2	10.12.2014 16:13	14	39	50	53,3	91,9	497,7	15	00:20:05	94/8/49%	29,4	7,7	123,4	5,80
WT20141126	4	10.12.2014 16:46	14	42	48,2	51,7	95,2	486,86	15	00:20:05	99/45/77%	27,1	7,3	120,7	5,43
WT20141126	1	10.12.2014 17:19	14	40	51	54,1	91,5	491,04	15	00:20:04	100/100/100%	24,2	7,8	107,2	5,91
WT20141126	4	10.12.2014 17:52	14	43	48,9	53,2	89,8	480,41	15	00:20:05	100/82/98%	29,8	7,3	132,3	5,92
WT20141126	2	10.12.2014 18:27	14	40	47	50,1	87,5	478,69	15	00:20:05	100/79/95%	24,8	7	97,8	5,73
WTbigvssmall20141126	4	11.12.2014 11:52	15	1	48,6	53,6	87,6	483,67	15	00:20:05	100/91/100%	26,2	7,6	120,6	6,12
WTbigvssmall20141126	4	11.12.2014 12:21	15	2	45,9	50,1	87,3	478,08	15	00:20:04	100/78/99%	24,3	7	100,9	5,74
WTbigvssmall20141126	4	11.12.2014 12:52	15	3	48	51,7	87	478,16	15	00:20:05	100/88/100%	25,7	7	104,4	5,94
WTbigvssmall20141126	4	11.12.2014 13:25	15	4	45,2	48,3	86,7	474,64	15	00:20:05	99/7/90%	25	6,8	103,4	5,57
WTbigvssmall20141126	4	11.12.2014 13:56	15	5	48,2	52,1	86,5	465,67	15	00:20:05	96/1/57%	25	7,3	104	6,02
WTbigvssmall20141126	4	11.12.2014 16:25	15	6	47,1	50,6	87,5	462,47	15	00:20:05	100/29/98%	24,7	7,4	103,5	5,78
WT20141126	1	14.12.2014 10:44	18	41	45,8	49,9	74,5	466,47	15	00:20:20	100/100/100%	23,6	7,5	105	6,70
WT20141126	2	14.12.2014 11:20	18	41	45,6	50	90,2	475,76	15	00:20:05	100/99/100%	23,3	7,3	99,7	5,54
WT20141126	4	14.12.2014 11:59	18	44	47,9	51,2	76,2	484,43	15	00:20:04	100/75/75%	32,6	7,3	137,9	6,72
WT20141126	4	14.12.2014 12:31	18	45	50,5	55	79,5	465,57	15	00:20:05	100/97/100%	30,7	7,2	129,2	6,92
WT20141126	1	14.12.2014 12:57	18	42	49,5	52,5	78,1	463,71	15	00:20:05	100/100/100%	35,2	7,3	154,3	6,72
WT20141126	2	14.12.2014 13:48	18	42	46	50,2	69,3	464,83	15	00:20:05	100/99/100%	28,6	8	130,8	7,24
WT20141126	2	14.12.2014 14:26	18	43	47,1	50,5	75	480,01	15	00:20:05	100/95/100%	28	7	111,7	6,73
WT20141126	1	14.12.2014 15:00	18	43	45,6	48,8	88,8	457,64	15	00:20:04	100/100/100%	26,1	7,1	102	5,50
WT20141126	4	14.12.2014 15:33	18	46	47,6	50,9	80	478,7	15	00:20:05	100/59/99%	29,3	7,3	121,2	6,36

WT20141126	1	14.12.2014 16:03	18	44	45,8	48,9	82,1	475,04	15	00:20:05	100/100/100%	23,1	7	92,2	5,96
WT20141126	4	14.12.2014 16:40	18	47	48	52,1	85,6	483,26	15	00:20:05	100/77/99%	26,3	7,6	113,8	6,09
WT20141126	2	14.12.2014 17:09	18	44	50,1	53,1	75	492,33	15	00:20:05	100/97/100%	29,8	7,3	135,1	7,08
WT20141126	1	17.12.2014 12:10	21	45	46,2	51,9	87,2	467,85	15	00:20:05	100/100/100%	22,8	8,2	108,6	5,95
WT20141126	4	17.12.2014 13:14	21	48	47,9	52	79,7	478,85	15	00:20:05	99/44/93%	25,6	7,4	108	6,52
WT20141126	2	17.12.2014 13:57	21	45	46,2	49,3	58,5	471,87	18	00:20:04	100/100/100%	29,2	7,3	139,3	8,43
WT20141126	2	17.12.2014 14:31	21	46	48,5	52,2	62,6	469,55	18	00:20:05	100/84/99%	32,8	7,7	158,7	8,34
WT20141126	4	17.12.2014 15:04	21	49	48,3	51,6	65,4	450,74	15	00:20:04	100/96/99%	27	7,7	132,1	7,89
WT20141126	1	17.12.2014 15:31	21	46	48,5	51,7	66,8	456,11	15	00:20:05	100/100/100%	29,7	7,5	138	7,74
WT20141126	2	17.12.2014 16:18	21	47	46,8	50,5	65,1	461,23	15	00:20:05	100/95/100%	27,8	7,6	129,3	7,76
WT20141126	4	17.12.2014 17:26	21	50	49	53,2	65,4	471,65	15	00:20:05	100/92/100%	32,2	7,8	162,7	8,13
WT20141126	1	17.12.2014 17:56	21	47	44,3	48,2	65,8	462,56	15	00:20:05	100/100/100%	22,9	7,2	93,8	7,33
WT20141126	4	17.12.2014 18:59	21	51	48,6	53,6	73,6	480,06	15	00:20:05	100/93/100%	26,1	7,8	123,1	7,28
WT20141126	2	17.12.2014 19:29	21	48	46,4	50,1	72,6	481,97	15	00:20:07	100/97/100%	24,7	7,3	107,8	6,90
WT20141126	1	17.12.2014 20:02	21	48	46,5	50,7	72,9	481,96	15	00:20:05	100/100/100%	27,9	7,9	135,3	6,95
WTAB20141126	4	18.12.2014 13:45	22	1	48,5	53,3	68,2	489,02	15	00:25:20	93/71/71%	35,9	7,9	168	7,82
WTAB20141126	4	18.12.2014 14:25	22	2	47,1	51,2	68,2	463,53	15	00:25:22	98/66/79%	29,3	7,8	140,7	7,51
WTAB20141126	4	18.12.2014 15:06	22	3	46	50,1	68,2	462,83	15	00:25:20	99/59/71%	30,1	7,7	140,3	7,35
WTAB20141126	4	18.12.2014 17:08	22	4	58,1	63	68,2	451,77	15	00:25:16	100/71/82%	42,7	8,6	223,3	9,24
WTAB20141126	4	18.12.2014 17:54	22	5	51,8	55,4	68,2	479,64	15	00:25:16	95/68/73%	39,6	8	186	8,12
WTAB20141126	4	18.12.2014 18:34	22	6	55,7	60	68,2	464,22	15	00:25:17	93/63/74%	38,7	8,7	211,5	8,80
WTAB20141126	4	18.12.2014 19:14	22	7	49,3	55,1	68,2	464,5	15	00:25:16		34	8,2	169,7	8,08
AB20141103	1	11.11.2014 18:16	8	1	37,6	22	23,4	439,55	15	00:20:08	100/100/100%	25	7,9	103,6	9,40
AB20141103	4	11.11.2014 18:57	8	1	39,3	22	24,5	483,01	15	00:20:15	98/22/74%	28,2	7	102,9	8,98
AB20141103	2	11.11.2014 19:41	8	1	39,3	47,1	139,4	440,88	15	00:11:40	100/41/100%	24,9	7	92,3	3,38
AB20141103	2	12.11.2014 18:21	9	2	41,5	48,5	127,2	479,02	15	00:20:09	100/48/84%	23,9	6,3	85	3,81
AB20141103	4	12.11.2014 19:00	9	2	43,8	50	124,1	487,86	15	00:20:08	98/0/53%	38,4	7	110,9	4,03
AB20141103	2	13.11.2014 13:01	10	3	36,2	41,7	125,3	457,91	15	00:20:09	100/83/99%	23,9	6,4	81,2	3,33
AB20141103	1	13.11.2014 13:38	10	2	38,1	44	115,5	437,92	15	00:20:09	100/100/100%	23,6	6,3	85,9	3,81
AB20141103	4	13.11.2014 14:10	10	3	37,1	43,5	116	518,82	15	00:20:09	99/32/83%	25,2	6,8	92	3,75

AB20141103	2	13.11.2014 15:40	10	4	39,6	45,4	117,8	438,1	15	00:20:12	98/25/63%	24,7	6,5	89,5	3,85
AB20141103	4	13.11.2014 16:19	10	4	38,8	44,7	116,3	442,66	15	00:20:09	98/4/70%	25,4	6,5	87,8	3,84
AB20141103	1	13.11.2014 16:47	10	3	37	43	115,1	440,4	15	00:20:08	100/100/100%	22	6,4	73,1	3,74
AB20141103	2	14.11.2014 12:10	11	5	39,1	44,5	111,1	433,11	15	00:20:09	97/34/63%	21,9	7,1	82,1	4,01
AB20141103	4	14.11.2014 12:45	11	5	36,3	41,7	114,2	451,35	15	00:20:09	96/7/45%	23,3	6,5	78,1	3,65
AB20141103	1	14.11.2014 13:15	11	4	39,2	44,7	112,3	453,29	15	00:20:07	100/73/73%	25,5	6,1	78,5	3,98
AB20141103	4	14.11.2014 13:47	11	6	37,8	44,1	112,8	444,86	15	00:20:10	100/26/86%	24,2	6,3	82	3,91
AB20141103	2	14.11.2014 16:04	11	6	39,3	45,2	111,6	463,8	15	00:20:07	100/47/83%	32	7	90,9	4,05
AB20141103	1	14.11.2014 17:03	11	5	34	39,6	112,1	393,75	15	00:20:08	100/99/99%	22,6	5,8	68	3,53
AB20141103	1	14.11.2014 17:57	11	6	40	46,4	111,1	449,12	15	00:20:08	100/63/63%	22,5	7,2	79,8	4,18
AB20141103	4	14.11.2014 18:32	11	7	40,1	45,8	109,1	459,02	15	00:20:07	99/15/78%	25,8	6,6	88,7	4,20
AB20141103	2	14.11.2014 19:52	11	7	36,4	41,3	110,3	436,61	15	00:20:08	99/53/84%	22,9	6,1	78,5	3,74
AB20141103	4	15.11.2014 14:19	12	8	40,5	46,3	113,5	469,11	15	00:20:07	100/18/93%	27,7	6,7	107,2	4,08
AB20141103	1	15.11.2014 14:52	12	7	39,7	44,6	112,5	459,18	15	00:20:07	100/93/93%	24,5	6,1	82,4	3,96
AB20141103	2	15.11.2014 15:27	12	8	39,1	44,7	114,8	462,55	15	00:20:07	99/37/68%	24,8	6,8	85,7	3,89
AB20141103	4	15.11.2014 16:01	12	9	43,8	49,8	112,8	491,58	15	00:20:08	96/0/52%	28,6	7	106,7	4,41
AB20141103	2	15.11.2014 16:31	12	9	43,3	49,2	114,8	475,47	15	00:20:07	100/79/89%	27,3	6,6	99,9	4,29
AB20141103	1	15.11.2014 16:57	12	8	44,2	50	112,3	461,08	15	00:20:07	100/100/100%	26,5	7	96,7	4,45
AB20141103	1	17.11.2014 12:08	14	9	40,8	45	105,1	477,74	15	00:20:08	100/100/100%	26,4	7	106,6	4,28
AB20141103	4	17.11.2014 12:44	14	10	41,3	46,5	102,1	503,78	15	00:20:07	99/30/72%	26,2	6,4	95,3	4,55
AB20141103	2	17.11.2014 13:16	14	10	41,9	46,7	101,6	479,13	15	00:20:08	99/37/79%	26,6	6,6	95,5	4,60
AB20141103	1	17.11.2014 13:45	14	10	42,3	46,9	105,1	462,61	15	00:20:08	100/96/96%	23	6,6	83,6	4,46
AB20141103	2	17.11.2014 14:18	14	11	45	48,7	102,7	466,23	15	00:20:08	97/11/66%	27,2	6,5	91,6	4,74
AB20141103	4	17.11.2014 14:48	14	11	39,1	43,1	103,3	439,54	15	00:20:09	100/33/91%	28,1	6,5	98,2	4,17
AB20141103	1	17.11.2014 15:28	14	11	36,2	40	102,7	404,06	15	00:20:06	100/98/98%	30,8	6,5	106,7	3,89
AB20141103	2	17.11.2014 15:53	14	12	37,3	41,9	104,1	442,17	15	00:20:08	100/73/97%	28,8	7	107,2	4,02
AB20141103	1	17.11.2014 16:23	14	12	33	36,6	103,4	424,41	15	00:20:08	100/99/99%	28,3	6,9	98,2	3,54
AB20141103	2	17.11.2014 17:22	14	13	37,9	42,1	104,6	439,05	15	00:20:08	99/57/78%	21	7,5	83	4,02
AB20141103	4	17.11.2014 17:56	14	12	41,1	46,7	116,3	457,75	15	00:20:08	100/38/94%	29,7	7	115,1	4,02
AB20141103	4	17.11.2014 18:30	14	13	43,7	48,8	109,7	462,04	15	00:20:08	100/38/97%	31,6	7,1	127,9	4,45
AB20141103	2	18.11.2014 10:57	15	14	42	48,3	103,4	472,23	15	00:20:08	100/85/96%	23,8	7,6	96,8	4,67
AB20141103	4	18.11.2014 11:45	15	14	43,8	47,9	109,7	480,55	15	00:20:08	100/79/98%	34,6	7,4	139,8	4,37
AB20141103	1	18.11.2014 12:14	15	13	42,7	47,5	105	466,7	15	00:20:07	100/100/100%	36,2	7,1	141,4	4,52
AB20141103	2	18.11.2014 12:48	15	15	42,7	47,9	102	475,94	15	00:20:08	100/62/98%	28,5	7,6	120,4	4,70
AB20141103	1	18.11.2014 13:17	15	14	37,1	42,4	102	417,19	15	00:20:06	100/100/100%	29,7	6,8	109,6	4,16
AB20141103	4	18.11.2014 13:54	15	15	42,3	46,9	103,6	438,16	15	00:20:08	98/7/71%	28,1	7	103,7	4,53
AB20141103	1	18.11.2014 14:26	15	15	42,9	48,6	104,6	450,53	15	00:20:07	100/99/99%	29,4	7,8	114,8	4,65

AB20141103	2	18.11.2014 15:04	15	16	39,8	46,1	100	472,48	15	00:20:08	100/64/96%	34,3	7,6	141,8	4,61
AB20141103	1	18.11.2014 15:33	15	16	33,8	38,9	98,7	414,7	15	00:20:09	100/100/100%	27	6,6	101,4	3,94
AB20141103	2	18.11.2014 17:45	15	17	37,9	45,2	98,5	473,96	15	00:20:07	100/85/100%	27,2	7,2	102	4,59
AB20141103	4	18.11.2014 18:14	15	16	39,7	45,1	100,8	456,78	15	00:20:08	100/30/96%	26,9	7,7	111,2	4,47
AB20141103	4	18.11.2014 18:50	15	17	37,6	44,1	100,8	437,71	15	00:20:07	100/28/97%	27,6	7	105,7	4,38
AB20141103	4	19.11.2014 10:47	16	18	42,2	48,9	102	481,44	15	00:20:08	89/19/49%	29,5	8,1	135,2	4,79
AB20141103	2	19.11.2014 11:18	16	18	44,2	50,9	98,3	473,11	15	00:20:08	97/32/81%	28,3	7,9	121,5	5,18
AB20141103	1	19.11.2014 11:51	16	17	32,9	37,7	102,1	369,72	15	00:20:07	100/83/83%	20,9	7,3	77,3	3,69
AB20141103	2	19.11.2014 12:19	16	19	39	45,4	102,4	465,87	15	00:20:07	100/83/100%	27,7	7,8	115,9	4,43
AB20141103	1	19.11.2014 12:46	16	18	40,5	45,1	99,8	455,14	15	00:20:08	100/100/100%	26,3	7,9	111,2	4,52
AB20141103	4	19.11.2014 13:35	16	19	38,9	45,4	101	446,98	15	00:20:08	91/8/41%	27,8	8,1	123	4,50
AB20141103	2	19.11.2014 14:01	16	20	36,9	43	103,3	414,63	15	00:20:08	99/49/86%	27,3	7,2	103,8	4,16
AB20141103	4	19.11.2014 16:10	16	20	38,2	43,7	94,5	473,29	15	00:20:14	100/83/100%	32,3	7,5	136,6	4,62
AB20141103	1	19.11.2014 16:36	16	19	37,3	43,1	97	431,24	15	00:20:08	100/100/100%	28,7	6,7	102,2	4,44
AB20141103	4	19.11.2014 17:09	16	21	39,5	45,5	95,7	465,21	15	00:20:07	100/83/100%	29,5	7,3	122,2	4,75
AB20141103	1	19.11.2014 17:36	16	20	36,1	43	95,7	444,56	15	00:20:08	100/100/100%	23,4	7,7	97,5	4,49
AB20141103	2	19.11.2014 18:05	16	21	34,7	40,5	94,2	421,86	15	00:20:07	97/73/76%	29	6,7	105,9	4,30
AB20141103	1	20.11.2014 10:37	17	21	37,6	42,8	93,7	440,8	15	00:20:09	100/100/100%	33,8	7,2	131,8	4,57
AB20141103	4	20.11.2014 11:05	17	22	38,3	42,9	95,8	462,35	15	00:20:07	100/48/92%	27,4	7,4	107,7	4,48
AB20141103	2	20.11.2014 11:37	17	22	40,2	44,3	88,3	475,78	15	00:20:07	100/41/93%	23	7,3	90,2	5,02
AB20141103	1	20.11.2014 12:14	17	22	35,4	41,5	114,8	453,98	15	00:20:09	100/99/99%	26,5	7,5	106,5	3,61
AB20141103	4	20.11.2014 12:43	17	23	42,8	48,5	102,1	479,52	15	00:20:10	96/1/54%	29,3	7,3	111	4,75
AB20141103	2	20.11.2014 13:15	17	23	45,4	51,3	100	488,76	15	00:20:07	82/23/48%	29,4	7,6	117,9	5,13
AB20141103	1	20.11.2014 13:45	17	23	43,5	49,6	102,1	487,94	15	00:20:08	100/100/100%	25,7	8,2	108,8	4,86
AB20141103	4	20.11.2014 14:32	17	24	39,7	44,9	100,8	465,38	15	00:20:09	97/2/57%	27,7	7,1	104,1	4,45
AB20141103	2	20.11.2014 14:58	17	24	38,8	43,5	101	466,07	15	00:20:08	99/52/86%	28,6	7,5	118,4	4,31
AB20141103	1	20.11.2014 15:23	17	24	39,5	46,3	99,6	473,94	15	00:20:08	100/100/100%	35,9	7,9	167,8	4,65
AB20141103	2	20.11.2014 17:32	17	25	39,9	45,8	100,1	468	15	00:20:10	100/84/99%	31,3	7,4	130,7	4,58
AB20141103	4	20.11.2014 18:08	17	25	38,1	43,9	96,9	468,1	15	00:20:08	99/4/70%	27,3	7,7	110,9	4,53
AB20141103	1	20.11.2014 18:45	17	25	36	41,7	98,2	465,32	15	00:20:09	100/100/100%	27,7	7,1	111,1	4,25
AB20141103	1	21.11.2014 10:37	18	26	41,4	46,7	95,8	458,07	15	00:20:09	100/100/100%	32,3	7,6	128,2	4,87
AB20141103	4	21.11.2014 11:07	18	26	39,9	45,7	105,1	446,94	15	00:20:08	100/29/93%	34,6	7,8	142,2	4,35
AB20141103	2	21.11.2014 11:34	18	26	42,6	47,8	102,2	484,27	15	00:20:08	86/26/59%	27,9	8	119,6	4,68
AB20141103	2	21.11.2014 12:03	18	27	40,1	45,6	103,6	491,43	15	00:20:07	99/49/87%	29,4	7,4	115,2	4,40
AB20141103	1	21.11.2014 12:49	18	27	39,2	44,9	102,1	470,76	15	00:20:07	100/100/100%	22,1	8	94,4	4,40
AB20141103	4	21.11.2014 13:28	18	27	46,4	51,4	100,8	485,89	15	00:20:10	99/28/82%	34,2	8,2	152,4	5,10
AB20141103	4	21.11.2014 14:14	18	28	42,4	48,2	99,5	489,12	15	00:20:07	99/18/77%	27,5	7,9	112,9	4,84

AB20141103	2	21.11.2014 14:42	18	28	42,2	48	100,3	485,32	15	00:20:08	100/35/93%	29,7	7,5	124,1	4,79
AB20141103	1	21.11.2014 16:51	18	28	40	44,7	93,2	451,96	15	00:20:08	100/100/100%	37,5	7,9	176	4,80
AB20141103	1	21.11.2014 17:23	18	29	41,2	48,4	93,2	471,6	15	00:20:06	100/100/100%	26,4	7,9	121,6	5,19
AB20141103	4	21.11.2014 17:56	18	29	41,6	46,4	89,3	485,03	15	00:20:07	98/6/69%	32,2	7,4	135,3	5,20
AB20141103	2	21.11.2014 18:23	18	29	40,1	45,1	86,7	472,02	15	00:20:07	100/89/99%	27,9	7,5	122,6	5,20
AB20141103	4	22.11.2014 11:19	19	30	39,7	45,1	81,6	472,71	15	00:20:09	100/79/99%	31,9	7,7	140,1	5,53
AB20141103	1	22.11.2014 11:47	19	30	36,8	40,9	92,1	416,79	15	00:20:07	100/100/100%	24,9	7,7	112,4	4,44
AB20141103	2	22.11.2014 12:16	19	30	38,4	43,3	89,3	440,82	15	00:20:08	100/63/98%	24,3	7,5	103,5	4,85
AB20141103	1	22.11.2014 12:44	19	31	38,9	43,7	91,9	441,74	15	00:20:08	100/100/100%	31,7	7,7	141,1	4,76
AB20141103	2	22.11.2014 13:13	19	31	38,7	42,8	89,5	443,43	15	00:20:09	100/42/84%	22	7,8	88,2	4,78
AB20141103	4	22.11.2014 13:44	19	31	39,7	45,8	89,5	479	15	00:20:09	100/88/99%	29,8	7,9	133,5	5,12
AB20141103	1	22.11.2014 14:12	19	32	37,9	43,2	88,7	464,16	15	00:20:07	100/99/99%	20,4	7,7	86	4,87
AB20141103	4	22.11.2014 15:03	19	32	40	44,8	91,8	463,47	15	00:20:14	94/3/54%	26,7	7,6	113	4,88
AB20141103	2	22.11.2014 15:34	19	32	37,6	42,6	88,2	471,32	15	00:20:08	98/46/87%	30,1	7,8	124,4	4,83
AB20141103	4	22.11.2014 16:10	19	33	44,6	49,6	89,3	480,38	15	00:20:08	92/3/45%	31,9	8,2	138,6	5,55
AB20141103	2	22.11.2014 16:38	19	33	39	44,5	89,3	443,44	15	00:20:07	99/26/81%	25,7	7,3	95,8	4,98
AB20141103	1	22.11.2014 17:43	19	33	38	43,3	90	457,77	15	00:20:08	100/100/100%	27,2	8,4	120,6	4,81
AB20141103	1	23.11.2014 11:43	20	34	43	47	80,8	481,48	15	00:20:08	100/100/100%	34,5	7,7	157,5	5,82
AB20141103	4	23.11.2014 12:18	20	34	42,6	47,7	91,1	491,98	15	00:20:08	96/9/61%	31,6	7,6	132,3	5,24
AB20141103	2	23.11.2014 12:47	20	34	43,9	48,7	81,7	472,46	15	00:20:08	100/97/100%	36,2	7,5	149,4	5,96
AB20141103	1	23.11.2014 13:14	20	35	42,5	46,5	79,9	479,33	15	00:20:07	100/100/100%	32,6	7,5	140,5	5,82
AB20141103	2	23.11.2014 13:48	20	35	46,4	50,8	75,8	479,73	15	00:20:07	100/91/99%	32,6	8,1	151,9	6,70
AB20141103	4	23.11.2014 14:18	20	35	45	49,4	86,9	477,8	15	00:20:06	100/39/96%	33	7,9	149,1	5,68
AB20141103	1	23.11.2014 14:48	20	36	40,8	44,4	87,9	454,16	15	00:20:07	100/100/100%	22,6	7,6	88,6	5,05
AB20141103	2	23.11.2014 15:14	20	36	41,3	45,5	89,1	463,48	15	00:20:08	97/33/81%	27,7	7,8	117,8	5,11
AB20141103	4	23.11.2014 15:55	20	36	43,6	48,5	83,6	490,91	15	00:20:14	100/51/92%	36,5	7,4	140,5	5,80
AB20141103	4	23.11.2014 16:34	20	37	42	46,3	83,3	476,36	15	00:20:07	100/66/97%	33,7	7,6	143,8	5,56
AB20141103	2	23.11.2014 17:05	20	37	41,4	45,1	89,8	465,61	15	00:20:07	100/73/97%	31	7,3	124,3	5,02
AB20141103	1	23.11.2014 17:35	20	37	46	50,6	110,1	483,52	15	00:20:07	100/99/99%	29,1	8,5	131,5	4,60
AB20141103	2	24.11.2014 11:20	21	38	39,4	42,7	87,7	443,31	15	00:20:08	100/95/100%	32,1	7,4	125,8	4,87
AB20141103	1	24.11.2014 11:46	21	38	42,5	46,3	86,6	447,19	15	00:20:07	100/100/100%	25,7	8	112,2	5,35
AB20141103	4	24.11.2014 12:14	21	38	45,7	50	87,1	489,86	15	00:20:07	98/31/85%	30,2	8,1	135,1	5,74
AB20141103	2	24.11.2014 12:54	21	39	42,2	45,8	69	482,45	15	00:20:07	100/99/99%	29,7	7,8	134,1	6,64
AB20141103	4	24.11.2014 13:23	21	39	44,6	48,6	85,8	462,88	15	00:20:10	100/62/98%	31,1	7,6	127,9	5,66
AB20141103	1	24.11.2014 14:23	21	39	41,2	46,1	71,7	488,08	15	00:20:07	100/100/100%	28,2	7,4	129,5	6,43
AB20141103	1	24.11.2014 16:24	21	40	43,7	47,9	89,6	468,98	15	00:20:09	100/100/100%	28,2	7,7	126,1	5,35
AB20141103	2	24.11.2014 16:51	21	40	37,8	42	86,2	457,91	15	00:20:07	97/82/89%	28,7	7,4	126,4	4,87

AB20141103	4	24.11.2014 17:22	21	40	41,3	45,3	80,8	476,44	15	00:20:07	100/87/100%	36,1	7,6	158,8	5,61
AB20141103	2	24.11.2014 17:50	21	41	45,5	50,1	81,7	480,46	15	00:20:07	100/98/100%	32	8,1	150,2	6,13
AB20141103	4	24.11.2014 18:23	21	41	42,9	47,8	91,6	476	15	00:20:07	99/30/79%	31,1	8,2	137,3	5,22
AB20141103	1	24.11.2014 19:08	21	41	42,7	47,2	78,5	487,35	15	00:20:08	100/100/100%	36,5	7,9	175,4	6,01
AB20141103	1	25.11.2014 10:50	22	42	39,7	43,9	77,8	439,2	15	00:20:07	100/100/100%	27,9	7,6	116,7	5,64
AB20141103	4	25.11.2014 11:27	22	42	45,1	48,9	72,8	472,54	15	00:20:07	100/100/100%	29,6	8,2	149,1	6,72
AB20141103	2	25.11.2014 11:55	22	42	41	45,9	75,5	449,52	15	00:20:07	100/100/100%	33,3	7,1	138,5	6,08
AB20141103	2	25.11.2014 12:23	22	43	39,6	43,1	72,1	472,1	15	00:20:08	100/97/97%	33,7	6,9	133,2	5,98
AB20141103	1	25.11.2014 12:48	22	43	37,4	40,8	72,7	426,46	15	00:20:08	100/100/100%	27,4	7,5	125,5	5,61
AB20141103	4	25.11.2014 13:17	22	43	40,5	45,8	85,4	464,85	15	00:20:07	100/82/98%	34	7,4	140,4	5,36
AB20141103	1	25.11.2014 13:57	22	44	39,5	43,3	85,5	440,69	15	00:20:08	100/100/100%	30,6	7	119,4	5,06
AB20141103	4	25.11.2014 15:03	22	44	40,4	46,7	77,6	477,63	15	00:20:09	99/86/87%	37,5	7,4	156	6,02
AB20141103	2	25.11.2014 15:31	22	44	41,9	46,2	70,2	466,5	15	00:20:08	100/100/100%	26,5	7,3	117,7	6,58
AB20141103	4	25.11.2014 17:26	22	45	40,4	44,3	67,9	454,45	15	00:20:08	100/100/100%	34,7	7,6	172,9	6,52
AB20141103	2	25.11.2014 17:58	22	45	47,6	52,3	69	493,78	15	00:20:07	100/100/100%	37,4	8,6	211,9	7,58
AB20141103	1	25.11.2014 18:32	22	45	44,7	49,3	79,2	470,1	15	00:20:08	100/100/100%	34,9	7,6	166,2	6,22
AB20141103	2	26.11.2014 11:06	23	46	46,1	49,7	66,5	475,35	15	00:20:08	100/100/100%	37,5	7,5	169,6	7,47
AB20141103	4	26.11.2014 11:44	23	46	40,2	44	58,7	475,71	18	00:20:08	100/100/100%	35,9	6,9	148,9	7,50
AB20141103	1	26.11.2014 12:13	23	46	39,8	44	65,4	464,27	15	00:20:07	100/100/100%	27,8	6,9	116,3	6,73
AB20141103	1	26.11.2014 12:40	23	47	38,7	42,1	65,2	437,92	15	00:20:09	100/100/100%	34,8	6,8	137,9	6,46
AB20141103	4	26.11.2014 13:07	23	47	37,1	41,3	65,1	423,57	15	00:20:07	100/97/99%	28,9	7,2	127,3	6,34
AB20141103	2	26.11.2014 13:43	23	47	40,1	44,7	75,4	451,63	15	00:20:07	100/98/98%	33	7,5	142,1	5,93
AB20141103	1	26.11.2014 14:58	23	48	42,2	46,3	67,8	463,42	15	00:20:13	100/100/100%	32,8	6,8	132,1	6,83
AB20141103	2	26.11.2014 16:11	23	48	42,2	46	70,2	473,74	15	00:20:13	100/100/100%	30	7,5	147,7	6,55
AB20141103	4	26.11.2014 16:43	23	48	41,8	45,3	71,5	459,91	15	00:20:08	100/99/100%	33,3	7,3	157,8	6,34
AB20141103	2	26.11.2014 17:43	23	49	40,9	44,2	71,8	460,51	15	00:20:08	100/88/88%	29,9	7,2	141,1	6,16
AB20141103	4	26.11.2014 18:10	23	49	45,3	49,1	72,1	476,18	15	00:20:06	100/100/100%	32	7,9	168,5	6,81
AB20141103	1	26.11.2014 18:43	23	49	42,9	46,6	72,1	480,12	15	00:20:09	100/100/100%	32,6	7,8	166,2	6,46
ABpetri20141103	1	27.11.2014 11:12	24	50	41,2	45,6	69,2	472,75	15	00:20:08	100/100/100%	23,4	7,2	88,9	6,59
AB20141103	1	27.11.2014 11:42	24	50	40,3	44,4	69,3	464,5	15	00:20:08	100/100/100%	28	7,2	104,8	6,41
AB20141103	2	27.11.2014 12:09	24	50	39,4	43	67,9	452,33	15	00:20:07	94/82/86%	24,2	7	102,1	6,33
ABpetri20141103	2	27.11.2014 12:41	24	50	38,3	42,9	68,9	463,62	15	00:20:07	100/99/99%	22,9	6,6	86,2	6,23
ABpetri20141103	4	27.11.2014 13:09	24	50	44,3	48,8	67,1	466,81	15	00:20:07	100/90/99%	26,6	7,1	115	7,27
AB20141103	4	27.11.2014 13:37	24	50	41,8	45,3	67,8	450,26	15	00:20:07	100/100/100%	25,9	7,4	116,7	6,68

AB20141103	4	27.11.2014 15:13	24	51	46,3	50,6	69	485,56	15	00:20:13	100/99/100%	36,2	7,9	184,1	7,33
ABpetri20141103	4	27.11.2014 16:25	24	51	46	50,5	69,3	460,58	15	00:20:13	100/91/100%	32,6	7,6	154,1	7,29
ABpetri20141103	1	27.11.2014 16:58	24	51	46,2	49,5	68,9	464,28	15	00:20:07	100/100/100%	29,8	8,1	151,6	7,18
AB20141103	1	27.11.2014 17:35	24	51	49	53,2	68,2	474,08	15	00:20:08	100/100/100%	31,8	8,4	173,1	7,80
AB20141103	2	27.11.2014 18:06	24	51	38,4	41,8	69,1	445,24	15	00:20:08	100/98/100%	30	7	127,5	6,05
ABpetri20141103	2	27.11.2014 18:32	24	51	38	41,4	67,8	456,73	15	00:20:07	100/93/99%	29,8	6,6	115,9	6,11
ABpetri20141103	2	28.11.2014 10:53	25	52	37,2	40,9	68,8	454,06	15	00:20:08	99/22/72%	19,4	6,5	72,3	5,94
ABpetri20141103	1	28.11.2014 11:27	25	52	46,3	49,8	67,9	453,42	15	00:20:06	100/99/99%	30,6	7	133,6	7,33
AB20141103	1	28.11.2014 11:59	25	52	46,7	50,5	67,6	491,82	15	00:20:06	100/100/100%	27,8	7,4	132,5	7,47
AB20141103	2	28.11.2014 13:01	25	52	40,1	44,1	69,1	452,33	15	00:20:07	100/89/100%	19,3	7	76,7	6,38
AB20141103	4	28.11.2014 14:54	25	52	45	49,2	68,4	474,42	15	00:20:06	100/100/100%	34,3	7,5	160,3	7,19
ABpetri20141103	4	28.11.2014 15:28	25	52	42,4	46,2	68	454,73	15	00:20:06	99/52/92%	30,7	7,1	127,1	6,79
AB20141126	1	01.12.2014 11:03	5	1	42,4	47,1	133,2	472,19	15	00:20:08	100/100/100%	36,5	6	110,5	3,54
AB20141126	2	01.12.2014 11:31	5	1	42,3	46,6	133,7	468,41	15	00:20:07	100/26/30%	42	6,5	144,1	3,49
AB20141126	4	01.12.2014 11:58	5	1	43,2	47,1	132,9	452,79	15	00:20:06	99/10/95%	36,9	6,3	123,5	3,54
AB20141126	2	01.12.2014 12:31	5	2	41	45,8	132,9	483,48	15	00:20:08	50/0/0%	37,3	6,8	136,9	3,45
AB20141126	1	01.12.2014 13:00	5	2	41,8	45,2	134	473,64	15	00:20:08	100/100/100%	38,9	6	117,6	3,37
AB20141126	4	01.12.2014 13:37	5	2	41,9	45,7	132,7	473,86	15	00:20:07	100/26/99%	31	6,4	103,7	3,44
AB20141126	1	01.12.2014 15:31	5	3	42,5	45,3	132,1	469,97	15	00:20:11	100/100/100%	31,9	6,4	104,8	3,43
AB20141126	2	01.12.2014 15:58	5	3	41,1	45,8	133,6	460,51	15	00:20:07	100/5/60%	33,8	6,1	112,5	3,43
AB20141126	4	01.12.2014 16:27	5	3	41,3	45,4	132,1	456,89	15	00:20:07	100/38/98%	29,9	6,3	96,9	3,44
AB20141126	2	01.12.2014 17:01	5	4	41,2	45,2	132,1	449,76	15	00:20:07	93/0/0%	16,8	3,1	57,7	3,42
AB20141126	1	01.12.2014 17:31	5	4	44,9	48,4	130,6	475,34	15	00:20:07	100/100/100%	41,6	6,2	138,7	3,71
AB20141126	4	01.12.2014 17:59	5	4	42,6	46,8	133,4	477,33	15	00:20:07	99/0/93%	35,7	6,1	115,9	3,51
AB20141126	1	02.12.2014 11:13	6	5	42,7	46,4	126,4	477,19	15	00:20:08	100/100/100%	26,3	6,5	91,4	3,67
AB20141126	2	02.12.2014 11:39	6	5	43,7	47,7	127,6	456,68	15	00:20:08	100/43/100%	29,9	6,2	95,6	3,74
AB20141126	4	02.12.2014 12:04	6	5	41,3	46	127,7	470,91	15	00:20:07	99/13/84%	34,8	6,3	115,4	3,60
AB20141126	2	02.12.2014 12:35	6	6	40,9	44,9	126,6	454,22	15	00:20:08	100/84/99%	26,2	6,4	86,5	3,55
AB20141126	4	02.12.2014 13:09	6	6	43	47	126,6	478,1	15	00:20:07	91/0/60%	31,2	6,4	107,2	3,71
AB20141126	1	02.12.2014 13:35	6	6	42,8	47,1	127,9	472,13	15	00:20:08	100/100/100%	39,4	6,3	129,3	3,68
AB20141126	4	02.12.2014 15:29	6	7	43	47,3	125,2	476,61	15	00:20:14	99/17/88%	32,1	6,3	102,8	3,78

AB20141126	1	02.12.2014 15:58	6	7	42,4	46,9	126,1	467,79	15	00:20:07	100/100/100%	36,3	6,4	120,4	3,72
AB20141126	2	02.12.2014 16:23	6	7	43	46,6	127,3	453,03	15	00:20:08	100/4/100%	34,2	6,1	113,7	3,66
AB20141126	2	02.12.2014 16:48	6	8	43,8	47,6	127,8	461,25	15	00:20:06	100/69/99%	26,6	6,3	91,8	3,72
AB20141126	4	02.12.2014 17:24	6	8	41,2	46	126,3	470,72	15	00:20:06	93/0/69%	24,1	6,5	89,7	3,64
AB20141126	1	02.12.2014 17:49	6	8	43,8	48,1	127,8	477,69	15	00:20:07	100/100/100%	29,3	6,5	95,6	3,76
AB20141126	4	03.12.2014 11:11	7	9	41,7	45,1	119,9	470,97	15	00:20:07	100/62/99%	35,3	6,4	119,7	3,76
AB20141126	1	03.12.2014 11:40	7	9	40,9	44,9	119,9	455,4	15	00:20:06	100/100/100%	36	6,2	116,1	3,74
AB20141126	2	03.12.2014 12:05	7	9	41,6	45,4	119,9	461,92	15	00:20:06	99/46/97%	27,1	6,2	90,2	3,79
AB20141126	1	03.12.2014 12:48	7	10	40,4	44,2	121,3	462,42	15	00:20:07	100/100/100%	34,8	6,2	110,8	3,64
AB20141126	2	03.12.2014 13:34	7	10	41,1	45,6	119,9	452,57	15	00:20:08	97/0/77%	20,5	6,4	73,8	3,80
AB20141126	4	03.12.2014 14:45	7	10	41,9	46,6	120,3	471,58	15	00:20:11	99/13/66%	24,4	6,3	82,1	3,87
AB20141126	4	03.12.2014 15:39	7	11	40,7	44,7	119,3	462,96	15	00:20:07	98/4/90%	25	6,3	85,9	3,75
AB20141126	2	03.12.2014 16:07	7	11	41,6	46,2	118,9	458,89	15	00:20:08	100/85/99%	27,6	6,4	93,3	3,89
AB20141126	2	03.12.2014 16:44	7	12	41,1	45,1	118,7	458,93	15	00:20:07	100/40/100%	29,6	6,1	95,9	3,80
AB20141126	1	03.12.2014 17:12	7	11	42,2	45,5	118,3	460,98	15	00:20:08	100/100/100%	25,6	6,3	87,4	3,85
AB20141126	4	03.12.2014 17:43	7	12	40,3	44,2	120,6	461,38	15	00:20:07	97/26/84%	26,6	6,3	88,2	3,67
AB20141126	1	03.12.2014 18:09	7	12	43,5	48,2	119,2	473,4	15	00:20:06	100/100/100%	25	6,5	84,5	4,04
AB20141126	2	04.12.2014 11:34	8	13	41,2	45,3	117,6	482,75	15	00:20:07	100/6/85%	21,1	6,9	80,6	3,85
AB20141126	1	04.12.2014 11:59	8	13	41	44,8	117,4	481,26	15	00:20:07	100/100/100%	24,7	6,2	83,7	3,82
AB20141126	4	04.12.2014 12:28	8	13	44	48	119,9	481,63	15	00:20:07	99/47/85%	24,9	6,4	88,1	4,00
AB20141126	1	04.12.2014 12:57	8	14	44,4	48	118,2	480,23	15	00:20:07	100/100/100%	36,1	6,4	128,6	4,06
AB20141126	4	04.12.2014 13:24	8	14	43,9	47,9	119	481,48	15	00:20:08	99/23/61%	32,3	6,9	121,5	4,03
AB20141126	2	04.12.2014 13:50	8	14	45	49	116,8	495,54	15	00:20:07	99/30/93%	29,4	6,8	109,1	4,20
AB20141126	4	04.12.2014 14:33	8	15	42,1	46,8	118,7	477,79	15	00:20:20	100/41/98%	28	6,7	101,5	3,94
AB20141126	2	04.12.2014 16:08	8	15	44,6	48,5	119	512,82	15	00:20:12	100/77/96%	27,4	6,7	97,7	4,08
AB20141126	1	04.12.2014 16:40	8	15	42,2	46,5	117,7	477,58	15	00:20:08	100/100/100%	27,9	6,7	108,9	3,95
AB20141126	2	04.12.2014 17:10	8	16	45,1	49,6	117,1	485,76	15	00:20:07	100/93/100%	30,8	6,5	115,4	4,24
AB20141126	4	04.12.2014 17:46	8	16	42,9	46,5	116,5	474,88	15	00:20:07	100/79/100%	27,8	6,8	105,8	3,99
AB20141126	1	04.12.2014 18:12	8	16	42,8	47,1	118,8	479,61	15	00:20:07	100/100/100%	32,1	7	125,7	3,96
AB20141126	4	05.12.2014 11:09	9	17	42,4	46,2	111,3	482	15	00:20:07	100/46/97%	30,1	7	116,8	4,15
ABpetri20141126	4	05.12.2014 11:39	9	1	41,8	45,9	108,7	462,05	9	00:20:08	100/69/98%	25,5	6,4	91,2	4,22
AB20141126	1	05.12.2014 12:04	9	17	41,4	45,3	112,3	481,28	15	00:20:07	100/100/100%	22,6	6,4	82,1	4,03
AB20141126	2	05.12.2014 12:39	9	17	42,4	46	111,4	470,92	15	00:20:06	96/80/90%	27,2	6,9	104,6	4,13
AB20141126	1	05.12.2014 13:46	9	18	41,1	44,6	112,7	450,63	15	00:20:13	100/100/100%	28,6	6,6	101,3	3,96
AB20141126	2	05.12.2014 14:20	9	18	43,6	47,2	113,5	483,78	15	00:20:08	100/14/98%	25,4	7,1	94,7	4,16
AB20141126	4	05.12.2014 15:38	9	18	41,6	46,3	112,3	487,52	15	00:20:08	93/22/56%	26,4	6,8	96,4	4,12

ABpetri20141126	4	05.12.2014 16:03	9	2	43,6	47,5	108,4	462,83	9	00:20:08	100/45/97%	24,5	6,7	88,6	4,38
AB20141126	2	05.12.2014 16:35	9	19	44,8	48,1	113,6	487,72	15	00:20:08	100/93/100%	29,4	6,5	106,3	4,23
AB20141126	4	05.12.2014 17:15	9	19	42,9	46,4	111,1	471,35	15	00:20:08	100/72/99%		30,2	7,1	4,18
ABpetri20141126	4	05.12.2014 17:50	9	3	43,6	48	109,5	462,02	9	00:20:07	100/8/94%	26,4	6,4	91,9	4,38
AB20141126	1	05.12.2014 18:16	9	19	43,9	47,4	111	480,53	15	00:20:08	100/100/100%	23,2	6,8	87,5	4,27
AB20141126	4	05.12.2014 18:47	9	20	43,2	47,4	112,7	481,62	15	00:20:07	100/55/96%	27,4	7,1	109,1	4,21
ABpetri20141126	4	05.12.2014 19:23	9	4	45,1	48,8	109,9	457,01	9	00:20:07	100/47/87%	28	6,9	102,4	4,44
AB20141126	2	05.12.2014 19:53	9	20	46,1	49,9	111,5	498,82	15	00:20:07	99/50/88%	27,4	6,6	100,2	4,48
AB20141126	1	05.12.2014 20:21	9	20	43,4	46,6	107,3	490,15	15	00:20:07	100/96/96%	24	6,3	86,3	4,34
AB20141126	4	06.12.2014 11:27	10	21	47,3	50,3	100	504,77	15	00:20:06	100/65/96%	35	6,7	126,7	5,03
AB20141126	1	06.12.2014 11:57	10	21	43,3	47,2	99,6	444,28	15	00:20:08	100/100/100%	22,4	6,4	79,6	4,74
AB20141126	2	06.12.2014 12:31	10	21	41,6	45	108,7	472,79	15	00:20:07	99/78/95%	26,6	6,7	99	4,14
AB20141126	2	06.12.2014 13:01	10	22	45,1	49,1	106,5	503,12	15	00:20:07	100/70/99%	29,3	7,2	114,8	4,61
AB20141126	2	06.12.2014 13:31	10	23	44,3	48,4	107,6	495,7	15	00:20:09	100/61/99%	33,1	7,3	136	4,50
AB20141126	1	06.12.2014 13:56	10	22	41,6	45,4	105,5	442,53	15	00:20:08	100/97/97%	25,3	6,5	86,6	4,30
AB20141126	1	06.12.2014 14:31	10	23	43,7	46,7	107,4	484,19	15	00:20:07	100/99/99%	25,7	7,2	102,2	4,35
AB20141126	4	06.12.2014 14:59	10	22	46,1	49,7	101,4	497,32	15	00:20:06	88/28/57%	34,4	7,3	145,7	4,90
AB20141126	4	06.12.2014 15:43	10	23	44,4	47,4	102,2	461,74	15	00:20:12	99/53/82%	32,1	7	131,4	4,64
AB20141126	2	06.12.2014 16:20	10	24	44,9	48,6	103	471,81	15	00:20:06	100/56/93%	28,8	6,9	113,7	4,72
AB20141126	4	06.12.2014 16:50	10	24	44,6	47,7	97,8	476,54	15	00:20:06	94/23/64%	29	6,6	107,3	4,88
AB20141126	1	06.12.2014 17:18	10	24	43,8	48,8	95,1	491,24	15	00:20:08	100/100/100%	30,8	6,8	119,7	5,13
AB20141126	1	07.12.2014 11:50	11	25	44,6	48,4	100,9	464,49	15	00:20:07	100/93/93%	21	7,1	83,7	4,80
AB20141126	4	07.12.2014 12:19	11	25	42,8	45,8	93,5	487,47	15	00:20:08	100/88/99%	33,4	6,6	126,1	4,90
AB20141126	2	07.12.2014 12:47	11	25	44,5	48	93,2	493,31	15	00:20:06	100/96/98%	38,1	6,5	141	5,15
AB20141126	2	07.12.2014 13:29	11	26	47,1	50,7	95,6	486,56	15	00:20:06	100/93/97%	29,5	7,1	120,2	5,30
AB20141126	1	07.12.2014 13:56	11	26	45,9	49	94,5	487,41	15	00:20:06	100/100/100%	37,7	6,8	145,8	5,19
AB20141126	4	07.12.2014 14:25	11	26	43,4	46,7	91,9	473,33	15	00:20:08	100/78/96%	28,2	6,7	110,9	5,08
AB20141126	1	07.12.2014 14:56	11	27	39,7	42,7	90,6	456,92	15	00:20:07	100/100/100%	26,2	6,1	93,6	4,71
AB20141126	4	07.12.2014 15:28	11	27	41,3	44,6	89,3	469	15	00:20:08	98/88/94%	30	6,5	113,5	4,99
AB20141126	2	07.12.2014 16:13	11	27	44,5	46,9	97,2	463,97	15	00:20:11	100/96/100%	30	6,7	116,2	4,83
AB20141126	4	07.12.2014 16:48	11	28	45,6	49,3	94,2	479,78	15	00:20:07	99/20/69%	26,2	6,4	97,5	5,23
AB20141126	2	07.12.2014 17:26	11	28	40,6	44,2	101,7	482,46	15	00:20:07	100/98/100%	27,4	6,6	104,7	4,35
AB20141126	1	07.12.2014 17:56	11	28	48	50,8	97,1	489,25	15	00:20:07	100/100/100%	30,3	7	115,8	5,23
AB20141126	4	07.12.2014 18:24	11	29	44	47,2	97	487,97	15	00:20:06	100/69/90%	30,4	6,9	118,2	4,87

AB20141126	1	08.12.2014 11:28	12	29	45,9	48,6	94,4	481,31	15	00:20:07	100/90/90%	21,3	7,3	83,1	5,15
AB20141126	4	08.12.2014 11:58	12	30	43,5	46,7	97,1	490,05	15	00:20:06	100/93/99%	30,1	6,9	115	4,81
AB20141126	2	08.12.2014 12:29	12	29	45,3	48,7	91,9	487,07	15	00:20:07	100/98/100%	27,1	6,7	108,6	5,30
AB20141126	1	08.12.2014 12:55	12	30	43,1	47,1	91,9	458	15	00:20:08	100/100/100%	26,9	7	106,5	5,13
AB20141126	2	08.12.2014 13:28	12	30	42,8	45,9	96	496,41	15	00:20:07	100/97/100%	30,5	7,4	127,3	4,78
AB20141126	4	08.12.2014 13:59	12	31	47,4	50,9	97,1	492,64	15	00:20:07	99/29/80%	31,6	7,1	124,6	5,24
AB20141126	4	08.12.2014 14:39	12	32	45,6	48,8	90,8	484,12	15	00:20:07	100/52/93%	32,4	6,7	120,9	5,37
AB20141126	2	08.12.2014 15:06	12	31	46	50,3	90,6	487,12	15	00:20:07	100/78/99%	30,6	6,8	119	5,55
AB20141126	1	08.12.2014 15:35	12	31	46,8	50,9	88,3	490,39	15	00:20:07	100/100/100%	31	6,5	119,8	5,76
AB20141126	4	08.12.2014 16:01	12	33	44,2	47,6	88,6	472,44	15	00:20:07	100/76/94%	31,1	6,7	120,6	5,37
AB20141126	2	08.12.2014 16:59	12	32	42,6	46,2	90,6	485,36	15	00:20:07	99/87/97%	32,4	6,7	121	5,10
AB20141126	1	08.12.2014 17:26	12	32	41,7	45,8	87,1	460,88	15	00:20:14	100/100/100%	29,7	6,7	111,2	5,26
AB20141126	4	08.12.2014 17:53	12	34	44,6	48,8	86,2	473,89	15	00:20:08	99/53/91%	28	6,7	105,4	5,66
AB20141126	1	09.12.2014 11:19	13	33	45,6	48,6	95,6	473,9	15	00:20:08	100/97/97%	21,8	7,1	85	5,08
AB20141126	2	09.12.2014 11:52	13	33	45,6	48,7	96,9	484	15	00:20:07	100/92/93%	30,5	7	116,8	5,03
AB20141126	4	09.12.2014 12:23	13	35	46,2	50,2	89,6	485,26	15	00:20:07	100/87/96%	32,8	7,3	135,8	5,60
AB20141126	4	09.12.2014 12:55	13	36	46,4	50,2	85,1	488,74	15	00:20:08	100/98/100%	35	7,2	148,4	5,90
AB20141126	1	09.12.2014 13:27	13	34	48,2	51,4	82,4	489,14	15	00:20:06	100/100/100%	41,5	7	169,7	6,24
AB20141126	2	09.12.2014 13:55	13	34	44,1	48	83,6	491,33	15	00:20:07	100/98/99%	32,7	6,8	125,6	5,74
AB20141126	1	09.12.2014 16:10	13	35	46	49,9	91,6	484,24	15	00:20:07	100/100/100%	32,1	6,6	115,3	5,45
AB20141126	2	09.12.2014 16:40	13	35	44,1	47,7	90,3	492,13	15	00:20:08	100/87/91%	34,8	7,4	147,8	5,28
AB20141126	4	09.12.2014 17:10	13	37	46,3	48,8	85,5	487,43	15	00:20:07	99/46/85%	32,2	7,1	131,6	5,71
AB20141126	2	09.12.2014 17:39	13	36	45,7	48,6	84,6	492,11	15	00:20:07	100/98/100%	29,8	7,2	126,5	5,74
AB20141126	4	09.12.2014 18:16	13	38	41,1	44,7	87,2	479,89	15	00:20:07	99/50/73%	28,8	6,6	105	5,13
AB20141126	4	09.12.2014 18:49	13	39	42,1	45,6	84,3	487,66	15	00:20:07	100/93/99%	30,9	6,9	124,5	5,41
AB20141126	1	09.12.2014 19:30	13	36	44,6	48,5	85,5	468,23	15	00:20:07	100/90/90%	22,6	6,5	77,5	5,67
AB20141126	4	10.12.2014 11:42	14	40	47,5	50,2	86,2	492,24	15	00:20:09	100/99/100%	41,5	7	162,7	5,82
AB20141126	1	10.12.2014 12:10	14	37	42,3	45,1	84,6	472,21	15	00:20:07	100/99/99%	26,4	6,9	100,3	5,33
AB20141126	2	10.12.2014 12:37	14	37	44,4	47,6	84,2	480,46	15	00:20:07	100/70/96%	26,7	7,1	100,1	5,65
AB20141126	2	10.12.2014 13:03	14	38	47,5	51,1	85,1	487,79	15	00:20:08	100/98/98%	38,3	7,2	154,6	6,00
AB20141126	4	10.12.2014 13:36	14	41	44,9	47,5	81,9	492,65	15	00:20:08	100/85/96%	34,1	7	135,9	5,80
AB20141126	1	10.12.2014 14:16	14	38	48,6	51,6	84,2	488,29	15	00:20:07	100/100/100%	34,8	7,1	140,7	6,13
AB20141126	1	10.12.2014 15:42	14	39	45,3	48,3	85,7	472,26	15	00:20:11	100/100/100%	33,8	7	134,4	5,64
AB20141126	2	10.12.2014 16:12	14	39	43	45	89,4	484,61	15	00:20:11	100/96/100%	32	7,1	130,1	5,03
AB20141126	4	10.12.2014 16:42	14	42	41,8	45,2	90,6	489,32	15	00:20:06	100/85/100%	30,5	6,8	119,6	4,99
AB20141126	1	10.12.2014 17:19	14	40	41,9	46,1	102,1	487,98	15	00:20:11	100/100/100%	42,3	7,5	170,5	4,52
AB20141126	4	10.12.2014 17:52	14	43	42,8	46,4	93,1	491,54	15	00:20:06	100/76/97%	26,4	7	101,2	4,98

AB20141126	2	10.12.2014 18:30	14	40	44,2	48,6	93,3	488,41	15	00:20:10	100/81/94%	28,3	7	110,8	5,21
ABbigvssmall20141126	4	11.12.2014 11:49	15	1	48,5	51,4	97,2	469,42	15	00:20:14	100/42/96%	34,8	7,6	146,6	5,29
ABbigvssmall20141126	4	11.12.2014 12:26	15	2	51,4	53,9	96,9	486,05	15	00:20:08	87/10/27%	34,3	8	146,6	5,56
ABbigvssmall20141126	4	11.12.2014 12:59	15	3	52,9	57	98,3	481,77	15	00:20:08	94/17/58%	32,8	8,1	151,4	5,80
ABbigvssmall20141126	4	11.12.2014 13:34	15	4	55,3	58,6	95,8	479,78	15	00:20:06	95/30/61%	31,7	8,2	151,4	6,12
ABbigvssmall20141126	4	11.12.2014 15:49	15	5	50	53,9	97,3	484,97	15	00:20:11	95/50/71%	35,4	7,7	149,4	5,54
ABbigvssmall20141126	4	11.12.2014 16:23	15	6	48,5	53,2	96	485,15	15	00:20:15	99/15/79%	34,1	7,7	151	5,54
AB20141126	1	14.12.2014 11:00	18	44	45,9	49,5	89,4	481,64	15	00:20:09	100/100/100%	30,9	7,4	117,5	5,54
AB20141126	2	14.12.2014 11:32	18	41	44	46,4	82,3	480,62	15	00:20:10	100/100/100%	32,6	7,1	129	5,64
AB20141126	4	14.12.2014 12:04	18	44	42,8	45,8	77,9	493,11	15	00:20:14	100/91/91%	34	7	137	5,88
AB20141126	4	14.12.2014 12:41	18	45	42,9	45,3	76,6	492,19	15	00:20:07	100/98/100%	33,7	6,6	126,2	5,91
AB20141126	1	14.12.2014 13:27	18	45	43,5	46,7	87	493,15	15	00:20:06	100/100/100%	25,9	7,3	106,1	5,37
AB20141126	2	14.12.2014 13:56	18	42	46,2	49	75,5	493,27	15	00:20:13	91/75/75%	30	6,7	121,4	6,49
AB20141126	2	14.12.2014 14:29	18	43	45,9	49,2	69,6	492,73	15	00:20:10	100/100/100%	31,5	6,9	129,1	7,07
AB20141126	1	14.12.2014 15:01	18	46	50	52,9	68,9	482,17	15	00:20:07	100/100/100%	41,7	7,2	182,1	7,68
AB20141126	4	14.12.2014 15:32	18	46	46	48,7	72,8	488,39	15	00:20:08	100/98/100%	31	7	132,7	6,69
AB20141126	1	14.12.2014 16:01	18	47	45,8	48,6	75,5	478,31	15	00:20:07	100/100/100%	28	6,2	104,7	6,44
AB20141126	4	14.12.2014 16:41	18	47	43,6	46,6	68,9	493,75	15	00:20:06	100/99/100%	29,8	7	127,4	6,76
AB20141126	2	14.12.2014 17:10	18	44	46,3	49,9	69,2	483,48	15	00:20:07	100/97/100%	27,2	7,4	121,8	7,21
AB20141126	1	17.12.2014 12:23	21	48	43,4	45,9	81,8	474,73	15	00:20:07	100/100/100%	26,2	6,9	100,1	5,61
AB20141126	4	17.12.2014 13:14	21	48	49	52,9	73	477,26	15	00:20:07	100/95/99%	29	7,3	125,5	7,25
AB20141126	2	17.12.2014 13:55	21	45	43,8	46,9	73,3	483,65	15	00:20:07	100/100/100%	27,9	7,2	123,2	6,40
AB20141126	2	17.12.2014 14:29	21	46	45	47,3	75,5	477,97	15	00:20:08	100/99/100%	27,6	7,1	110,6	6,26
AB20141126	4	17.12.2014 15:03	21	49	49,2	52	68,4	481,62	15	00:20:07	100/99/100%	33,6	6,9	137,2	7,60
AB20141126	1	17.12.2014 15:30	21	49	44,7	46,9	66,9	446,12	15	00:20:07	100/98/98%	23,3	7,2	97,6	7,01
AB20141126	2	17.12.2014 16:16	21	47	47,6	50,2	66,8	488,64	15	00:20:11	100/97/100%	26,8	7,3	119,5	7,51
AB20141126	4	17.12.2014 17:25	21	50	46,1	50,3	66,9	481,14	15	00:20:12	96/52/67%	29,3	7,4	125,8	7,52
AB20141126	1	17.12.2014 17:54	21	50	47,1	49,6	66,3	432,24	15	00:20:07	100/80/80%	29,5	7,4	137,5	7,48
AB20141126	4	17.12.2014 19:12	21	51	46,1	48,6	61,6	482,4	18	00:20:07	100/93/95%	31,7	7,2	139,4	7,89
AB20141126	2	17.12.2014 19:44	21	48	46,6	50,1	66,9	480,07	18	00:20:07	96/90/92%	26,8	7,5	119,6	7,49
AB20141126	1	17.12.2014 20:11	21	51	45,7	48,3	66,4	477,01	15	00:20:08	100/100/100%	26,9	7,6	123,3	7,27

ABWT20141126	4	18.12.2014 18:04	22	1	47,7	51,8	67	445,85	15	00:25:06	98/70/83%	34,2	8	170,2	7,73
ABWT20141126	4	18.12.2014 18:43	22	2	48,8	51,8	67	453,19	15	00:25:35	97/52/73%	34,6	7,8	159,5	7,73

B.2 Influence of Arena Geometry

In previous experiments with zebrafish larvae, animals showed a strong tendency to stay close to the walls and move along the walls of the arena, a petri dish. Figure B.1 compares the distribution of animal coordinates in the arena during a 20 minute experiment for a petri dish and a watch glass. Both arenas had diameter of 8 to 9 centimeters. As can be seen in

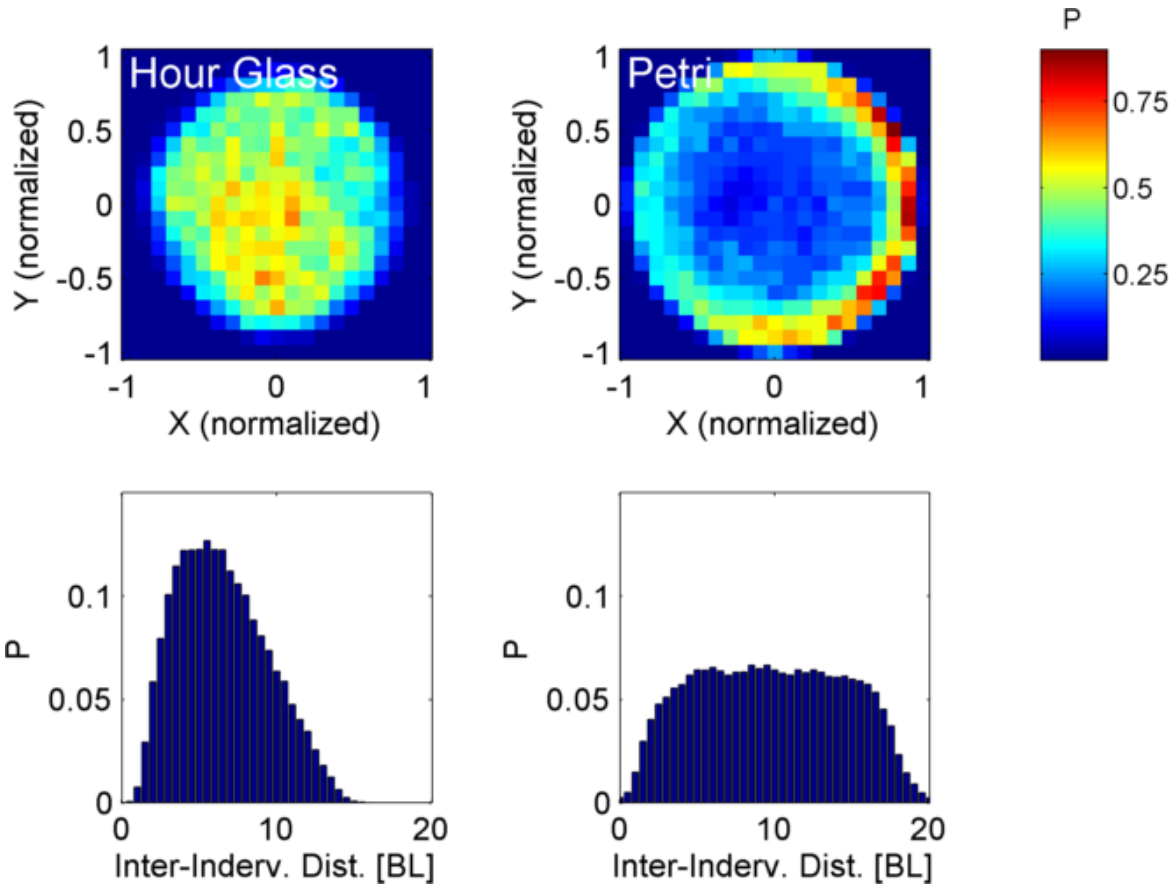

Fig. B.1 Distribution of animals in the arena for A) an hour glass and B) a petri dish of similar diameter (8-9cm). C) and D) show the reuslting distribution of inter-individual distances.

B.1, thigmotaxis strongly influences distribution of animals in the petri dish and interferes with social behavior, here given in terms of inter-individual distances.

While the influence of thigmotaxis can be eliminated by using watch glasses, a different type of border effect stays: When looking at turning behavior towards conspecifics, the probability increases the closer the focal individual comes to the border of the arena. This is the result of two combined factors: Firstly, the closer the wall is, the less likely is it that the focal will turn in the direction of the wall (Figure B.2c,d), the limiting case being the focal

touching the wall with the only possibility of turning towards the center of the arena. And secondly, the closer the wall, the more probable is it to find other conspecifics towards the center of the arena, simply because the space between the focal and the wall is very limited. As a result, an individual close to the border of the arena will turn towards its conspecifics with a higher probability than an individual at the center (Figure B.2a and b).

B.3 Methods for Trajectory Smoothing

When extracting coordinates of animals from video recordings, the results will never be exact due to a number of possible sources of noise (e.g., photo sensor, fluctuations of light intensity, segmentation). In order to get an idea of the influence of noise on behavioral analysis and of the importance of filtering or smoothing the data, Figure B.3 presents results obtained from recordings of an animal which stayed immobile troughout the 20 min of the experiment.

B.3.1 Assessment of Noise

B.3.2 Moving Average Smoothing

Moving average smoothing is a popular method for data smoothing due to its simplicity and robustness. It calculates the coordinate of an individual in a frame as the mean from a total number of *span* frames previous and subsequent to this frame. In practice, a trajectory obtained by *idTracker* is split into its x- and y- coordinate, which are then smoothed separately using the MATLAB® function *smooth* with parameters *span*=30 frames and *method* = 'moving'. The parameter value for *span* was chosen manually, by overlaying the resulting trajectory on top of the original video and making sure that coordinates taken from the smoothed trajectory do not deviate excessively from those taken from the original trajectory.

B.3.3 Adaptive Cubic Spline Smoothing

Cubic splines are another widely used tool for data smoothing and interpolation. For interpolation, polynomials of degree 3 (for cubic splines) are used to connect data points ('knots') under the conditions that both the 1st and 2nd derivatives of two polynomials meeting in a point do match (i.e., both derivatives of the curve are continuous) while maximizing the smoothness of the curve. For data smoothing, the knots do not have to coincide exactly with the data points, but are allowed to deviate from the original data points

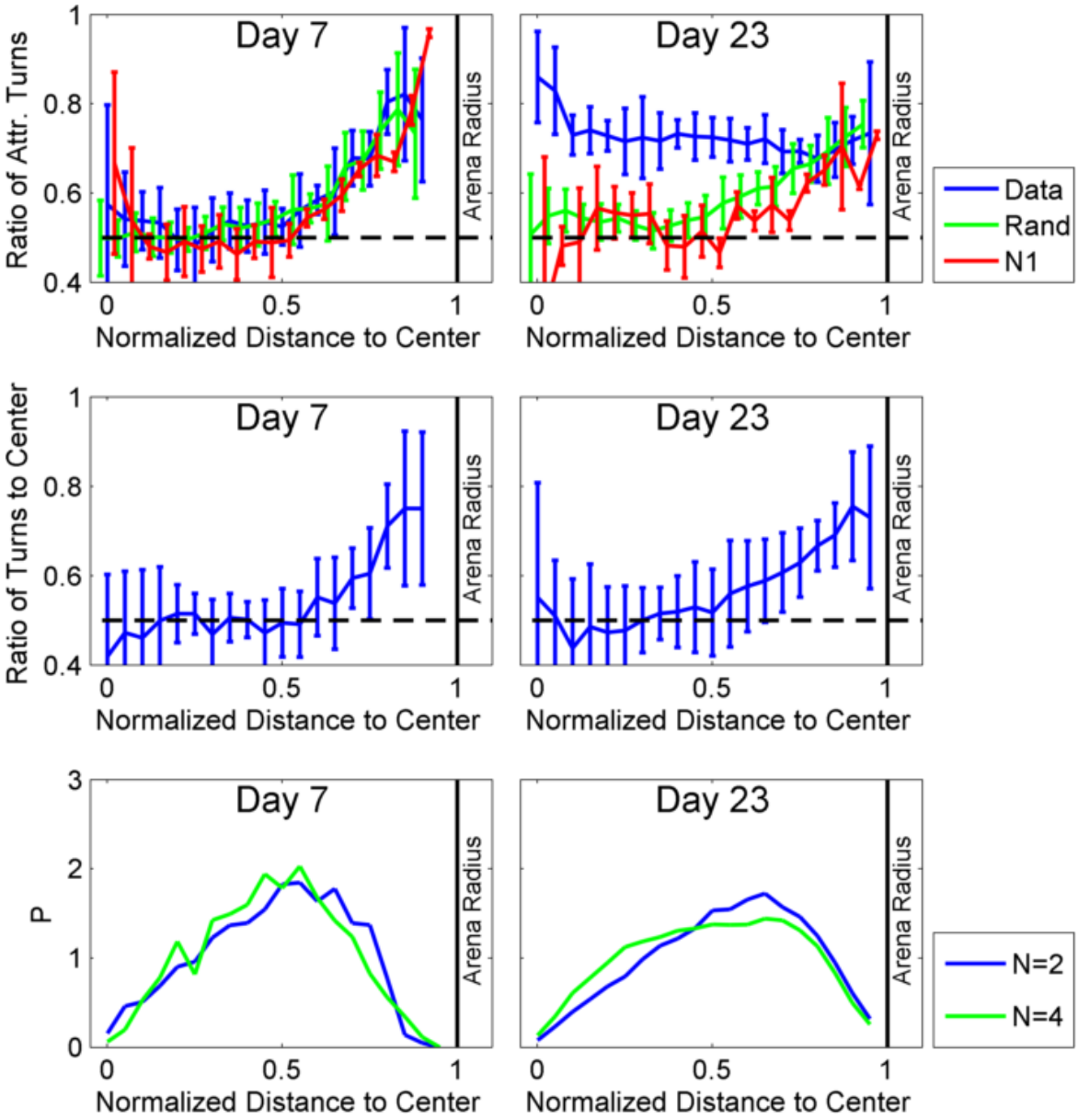

Fig. B.2 *Influence of proximity of arena borders on turning behavior* A), B) The ratio of turns towards a conspecific versus the distance of the focal from the center of the arena for an an experiment with zebrafish of age 7 and 23 dpf (blue). In addition, controls from randomization (green) and from joining trajectories of animals which were recorded separately (red) are shown. For comparison, C) and D) show the ratio of turns towards the center, without taking into account conspecifics. D) and E) show the probability of finding an animal at a certain radius from the center for groups of 2 (blue) and 4 (green) animals.

by a certain tolerance. The result is the smoothest function which lies within the given overall tolerance from the given data points.

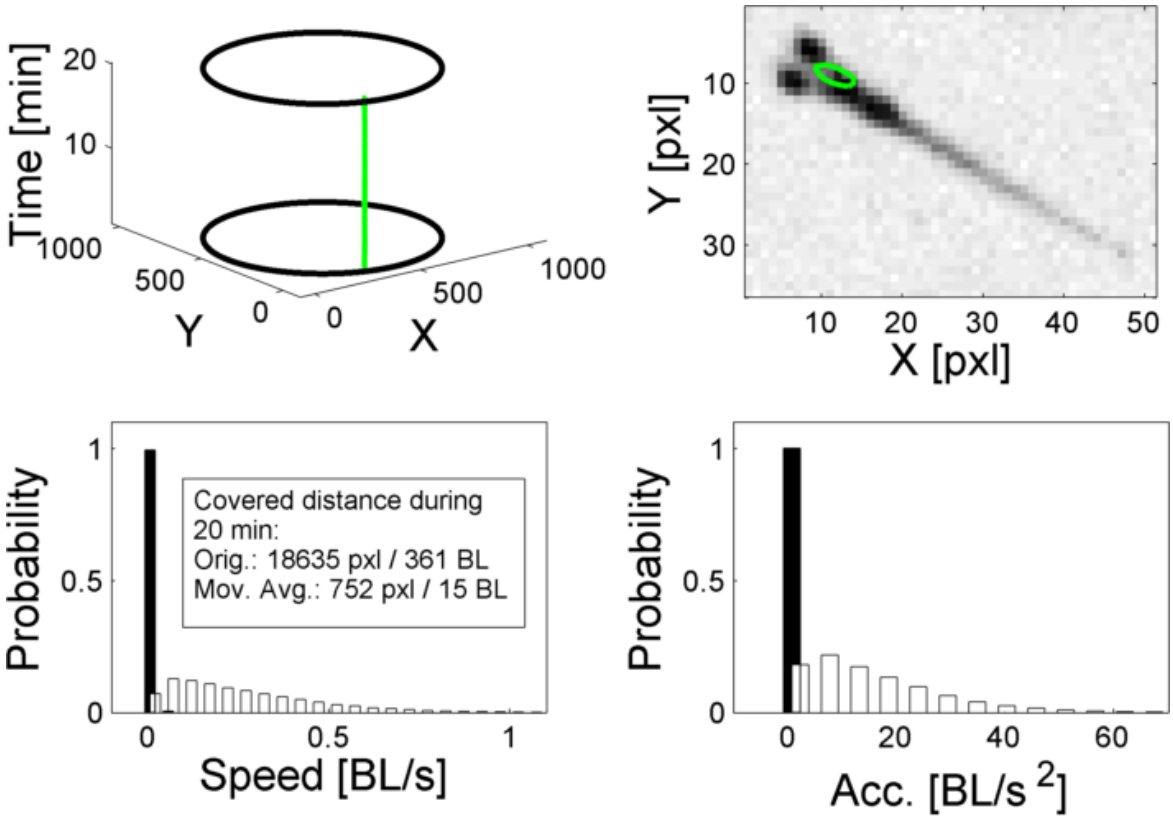

Fig. B.3 *Assesment of Noise* A) Tracjectory of an immobile animal B) Image of the immobile zebrafish larva (Blow-up from one example frame). The coordinates obtained for the 20 min of the experiment vary inside the region marked by a green circle. C) and D): Probability distribution of the speed and acceleration obtained from the original trajectory (white) and the smoothed trajectory (black).

Common implementations like the MATLAB® function *spaps* which follows the approach of C. Reinsch (C. Reinsch, "Smoothing by spline functions", Numer. Math. 10 (1967), 177–183.), calculate the distance between the function and the data by

$$E(f) = \sum_{j=1}^{n} w_j |(y_j - f(x_j))| \tag{B.1}$$

where w are additional weig!htbs which can be assigned to each data point, n is the number of data points, x are the data sites, and y the data values at the given sites. The roughness measure to be minimized is

$$F(f'') = \int_{\min(x)}^{\max(x)} \lambda(t) |f''(t)| dt \tag{B.2}$$

For our calculations we choose the roughness measure weig!htb $\lambda = 1$ and weig!htbs $w_j = 1$.

Instead of considering the sum of distances in Eq. B.1, for our purposes it is more convenient to control the maximum distance between each single coordinate of the original trajectory and its smoothed counterpart. This way we can not only avoid that the smoothed trajectory deviates unrealistically from the original at any data site, but also can we introduce a smoothing parameter which is linked to a length scale rather than to an arbitrary degree of smoothness. The advantage of the latter is that it makes it easier to interpret the smoothing parameter in terms of length scales observable in the experiments, especially body length. Therefore we developed an algorithm which adaptively matches the desired limit for any given pair of original and smoothed coordinate to the appropriate overall tolerance as required by Eq. B.1.

To do so, in a first step, a smoothing spline is calculated separately for the x- and y-coordinates of the trajectory using the built-in MATLAB® function *spaps* with an initial parameter *tol* = 30. Then the distance between every coordinate of the smoothed trajectory and the corresponding coordinate of the original trajectory is calculated. If more than 1% of the distances are greater than the chosen goal, set by parameter *FinalTolerance*, the initial tolerance is multiplied by 0.5, and smoothing is applied with this new tolerance. If 99% of the distances are smaller than *FinalTolerance* - *TolWidth*, where *TolWidth* is set to $0.1 * FinalTolerance$, the initial tolerance is multiplied by 2, and again smoothing is applied. Only if the 99th percentile of the distances lies between *FinalTolerance* - *TolWidth* and *FinalTolerance*, the algorithm stops (Fig. B.4).

For the following analysis we used a parameter value of *FinalTolerance* = 30 pixels when cubic spline smoothing was applied, which is a about half the body length of individuals in the experiments ($\approx$ 50 pixel).

Note that in applications where the 2nd derivative is of interest, a quintic spline smoothing can be more appropriate, because it guarantees that f'' itself is a continuous function (being itself a cubic spline), while for cubic splines the resulting acceleration curve will be a series of connected line segments. Nonetheless, this is rather a concern when one is interested in interpolated values not situated at the knots/data sites, but between them. However, we calculate velocity and acceleration values only at the given data sites (i.e., for each frame of the video), and see that our results vary little when comparing cubic and quintic splines, while computation time increases heavily.

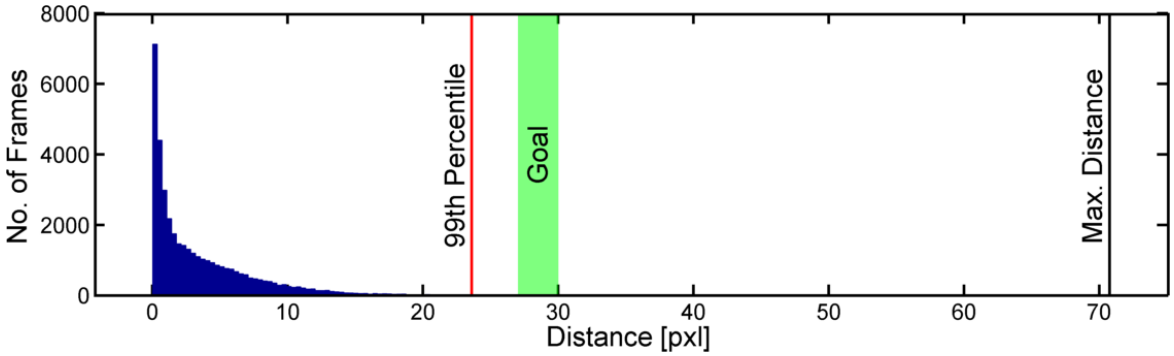

Fig. B.4 *Automatic Selection of overall tolerance for spline smoothing* Example step in the algorithm used to determine the parameter for spline smoothing which gives the desired spatial tolerance. The distribution of distances between the original and the smoothed trajectory is calculated. If the 99th percentile of the distribution lies outside the defined goal, the smoothing parameter is adjusted accordingly and the smoothing is repeated until the goal is reached.

From Fig. B.5 one can easily see that the absolute values of acceleration (as well as velocities, not in the figure), and thus also of the acceleration perpendicular to the direction of movement ('turning'), strongly depend on the method and degree of smoothing.

As a comparison between different methods of smoothing, Figure B.8 repeats the results shown in Figure 2.5 but for adaptive spline smoothing with a tolerance of 30 BL.

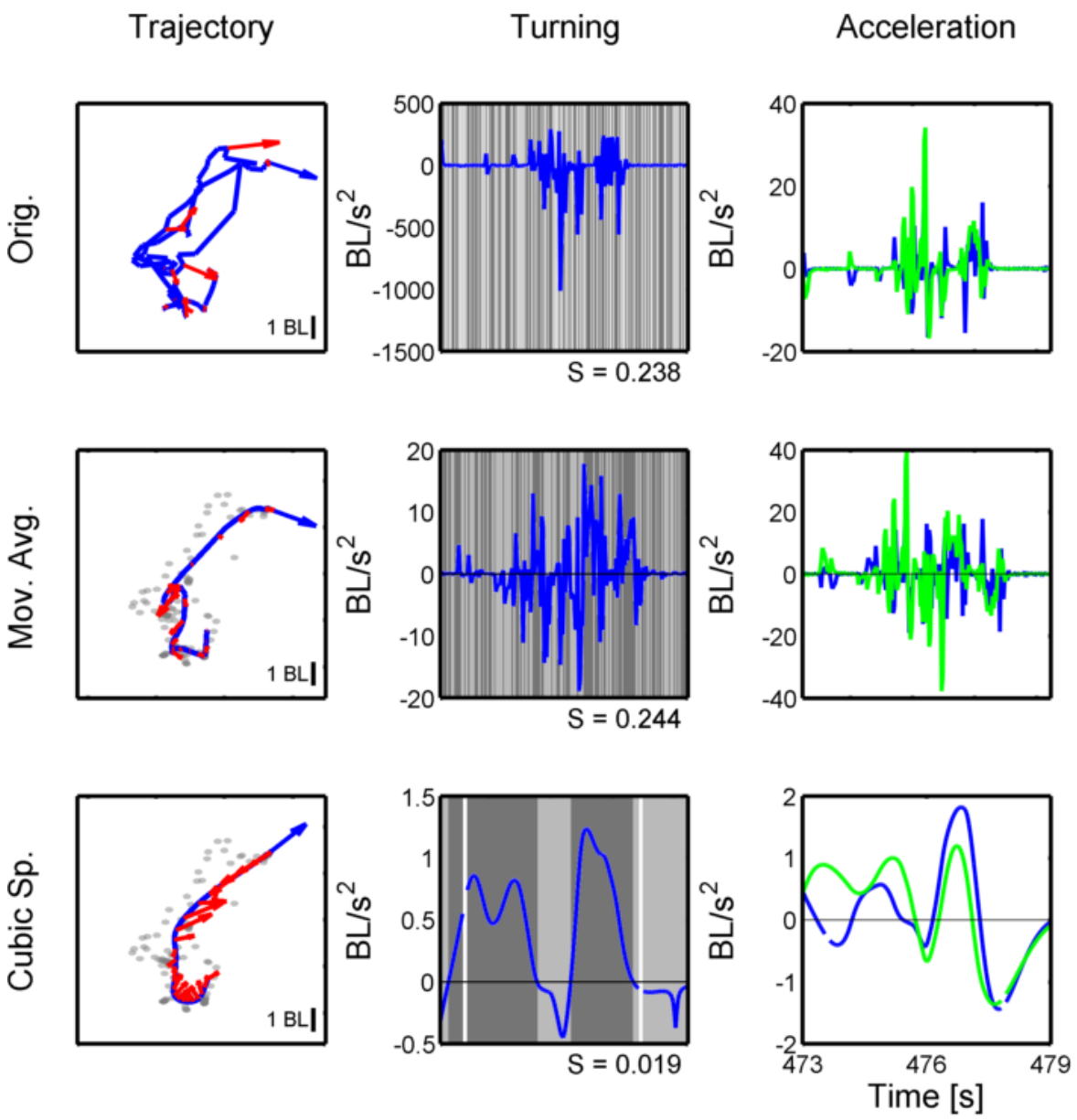

Fig. B.5 *Overview of Methods for Trajectory Smoothing* 1st row: No smoothing. 2nd row: Moving average smoothing with a window width of 30 frames, and 3rd row: Adaptive cubic spline smoothing (tolerance 30 pxl). 1st column: Trajectory (blue) after smoothing, original coordinates (gray dots) and acceleration vectors at a selection of coordinates (red arrows). 2nd column: Turning strength (component of the acceleration vector perpendicular to the direction of movement). The ratio of direction changes S is given below each figure. 3rd column: Forward acceleration/deceleration (component of the acceleration vector parallel to the direction of movement).

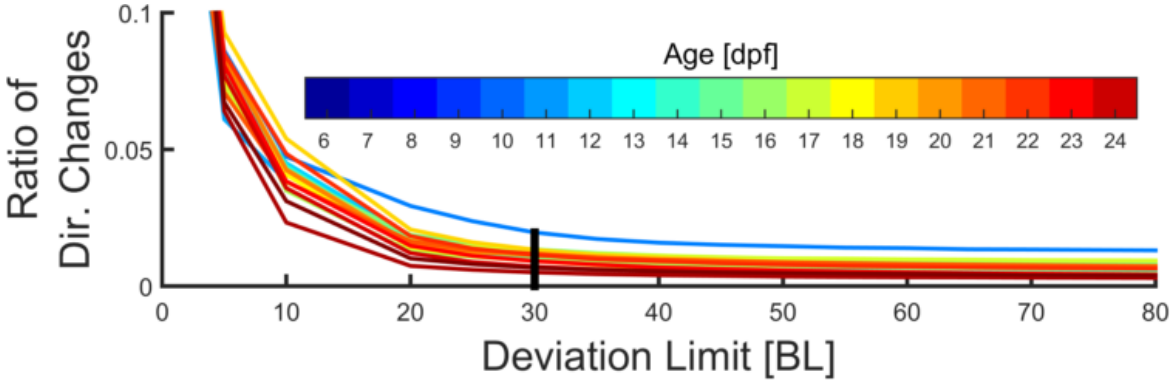

Fig. B.6 *Selection of Parameter for Adaptive Spline Smoothing* The ratio of changes of directions plotted versus the tolerance used for adaptive spline smoohing. Each colored line corresponds to the average over trajectories of given age. A vertical black line marks the parameter value which was finally used in our analysis.

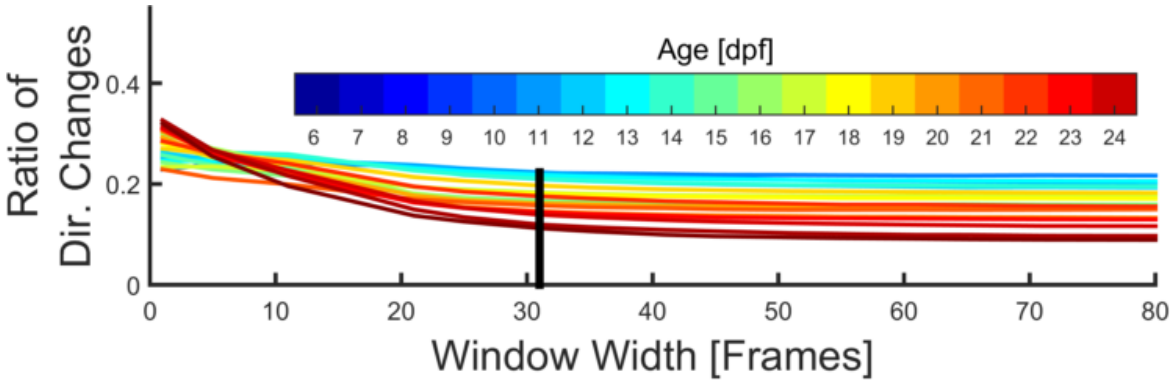

Fig. B.7 *Selection of Parameter for Moving Average Smoothing* The ratio of changes of directions plotted versus the tolerance used for moving average smoohing. Each colored line corresponds to the average over trajectories of given age. A vertical black line marks the parameter value which was finally used in our analysis.

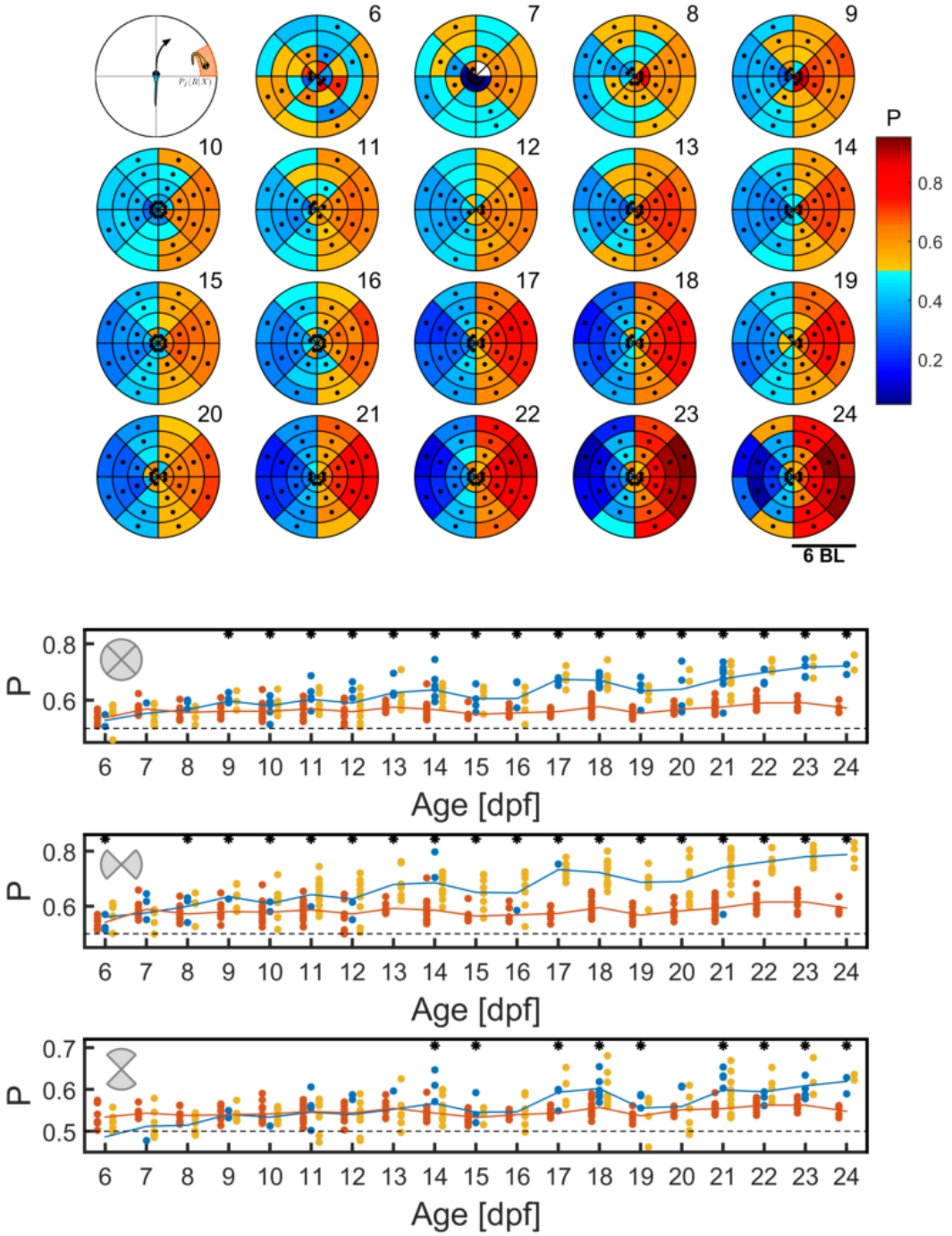

Fig. B.8 *Turning Behavior, Spline Smoothing* See Figure 2.5 for comparison with results from moving average smoothing. In the coordinate system with the focal at the origin and the vertical axis given by its direction of movement, a probability > 0.5 (< 0.5) of turning right for a given neighbor position is indicated by warm (cold) colors. B) Probability of turning right averaged over all pairs of the same age. C)-E) Probability of the focal turning towards the side of the neighbor, averaged over all pairs of the same age (blue solid line, stars indicate statistical significance) and for each pair of individuals (blue dots: Significantly different from randomized controls (Monte Carlo, $p<0.05$), yellow dots: No significant difference), C) taking into account all neighbors, D) only neighbors on the left/right, and E) only at the front/back. Orange dots/lines represent results from randomized controls.

B.4 Statistical Significance of Maps

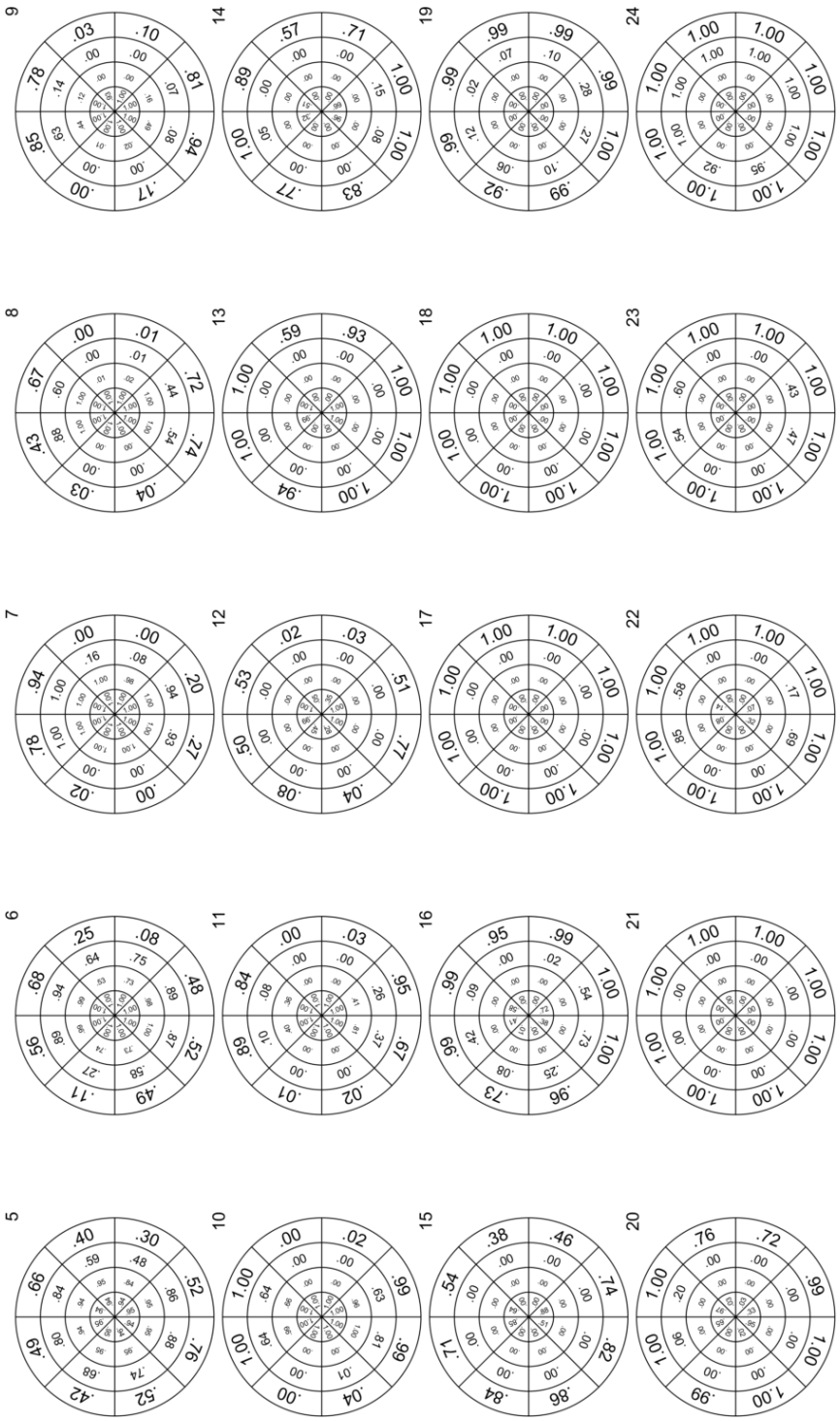

Fig. B.9 *Statistical Significance of Relative Positions* P-values for the maps shown in Figure 2.1F

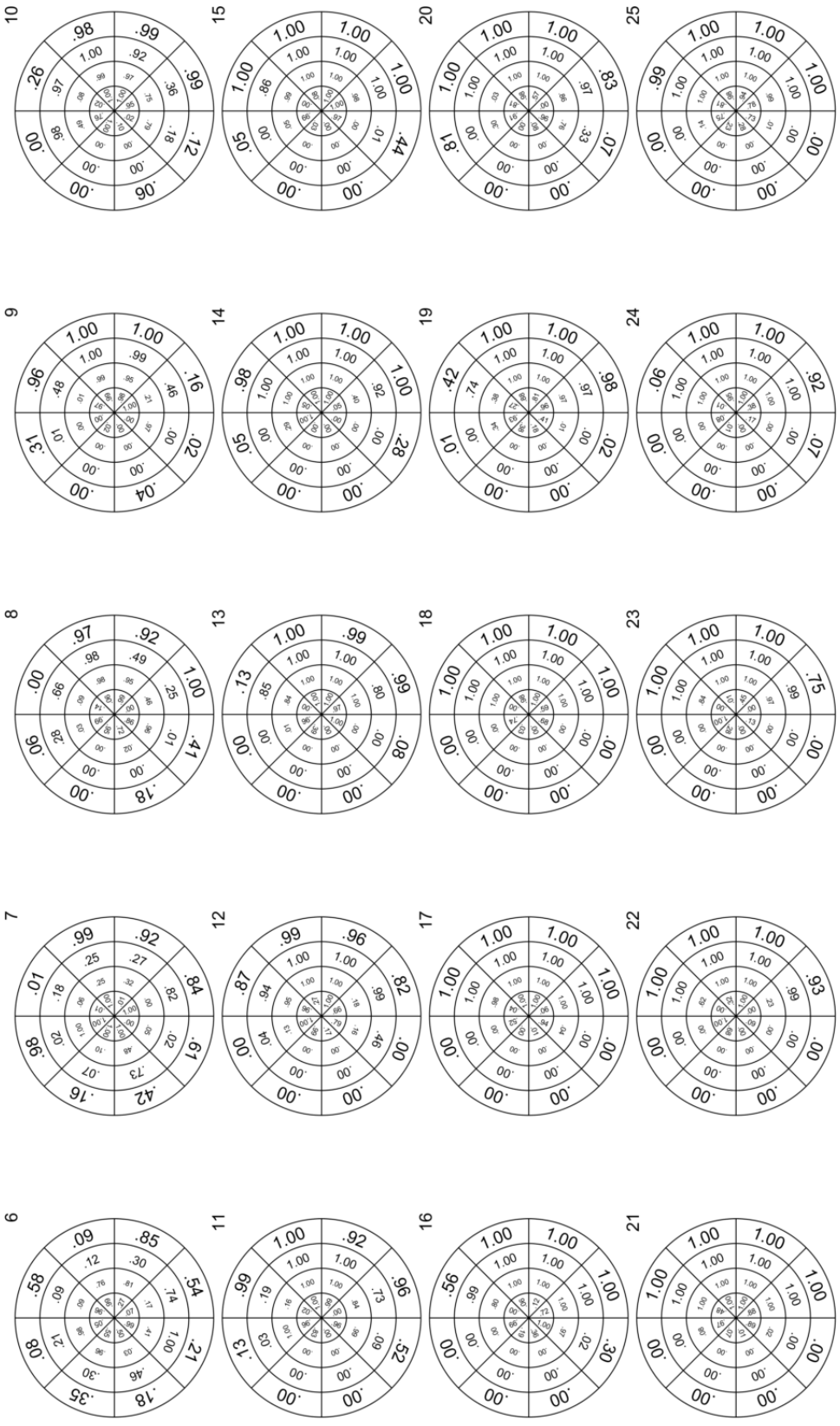

Fig. B.10 *Statistical Significance of Turning Behavior* P-values obtained from a right-tailed Monte Carlo test for the maps shown in Figure 2.5F

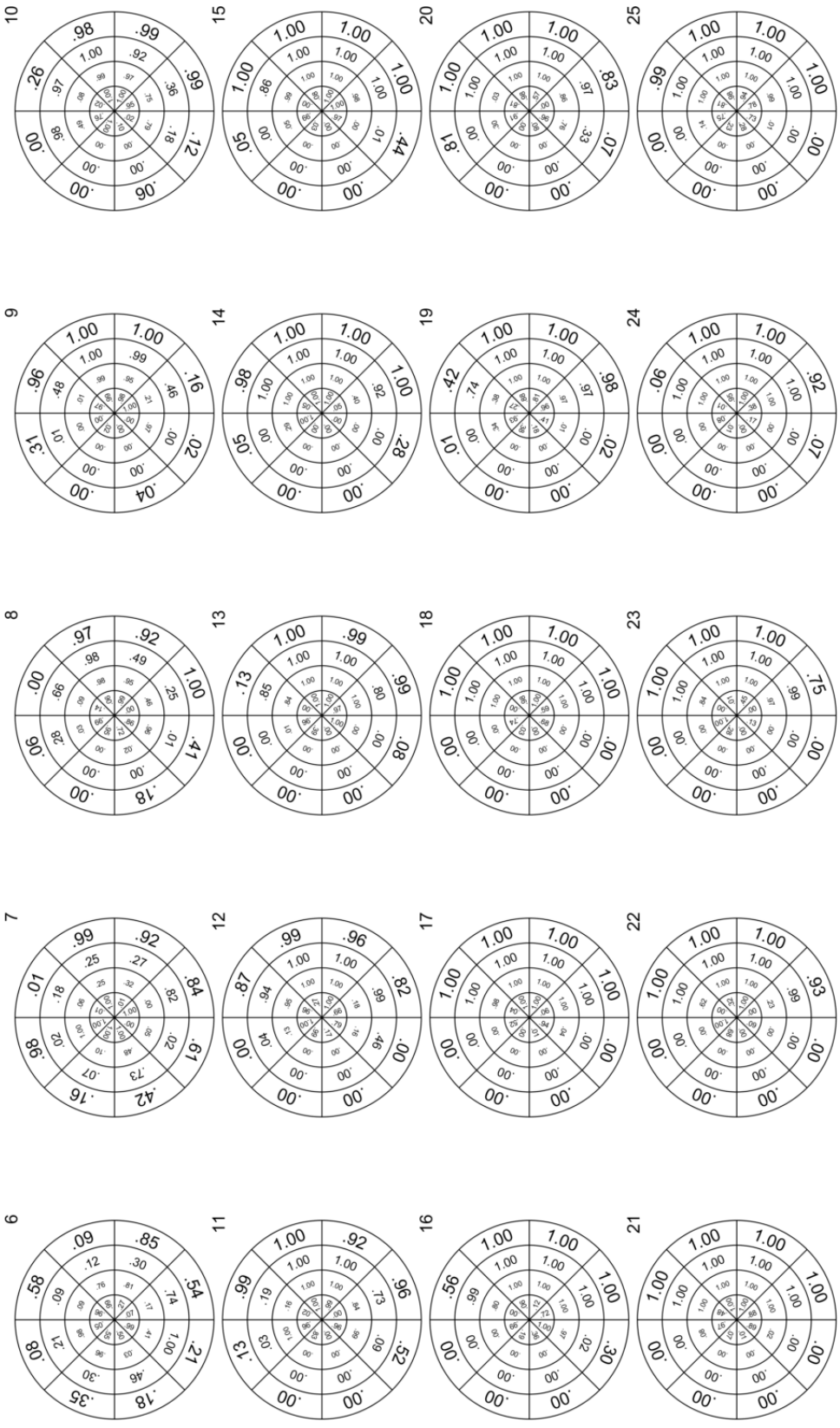

Fig. B.11 *Statistical Significance of Turning Behavior* P-values obtained from a left-tailed Monte Carlo test for the maps shown in Figure 2.5F

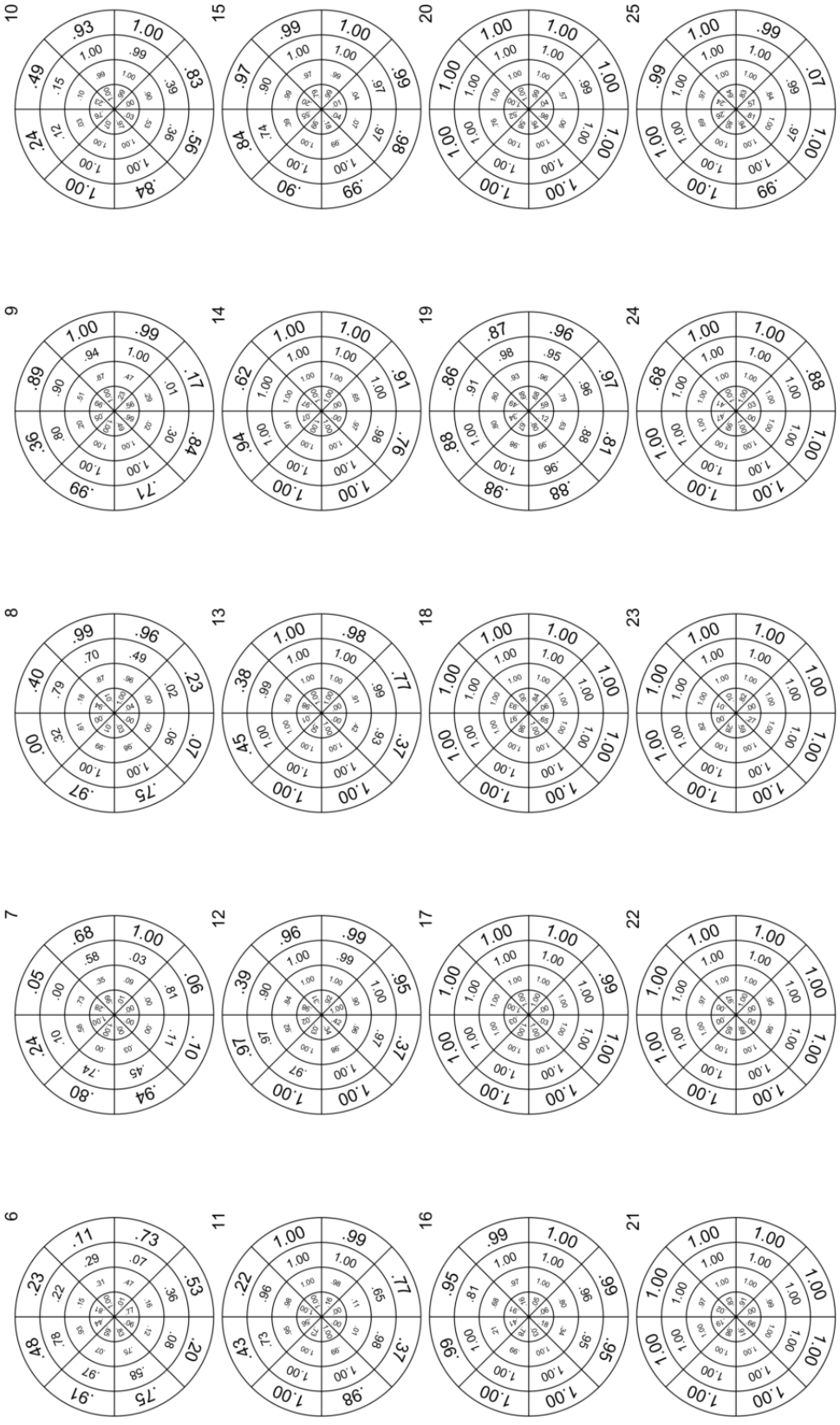

Fig. B.12 *Statistical Significance of Acceleration Behavior* P-values obtained from a right-tailed Monte Carlo test for the maps shown in Figure 2.6F

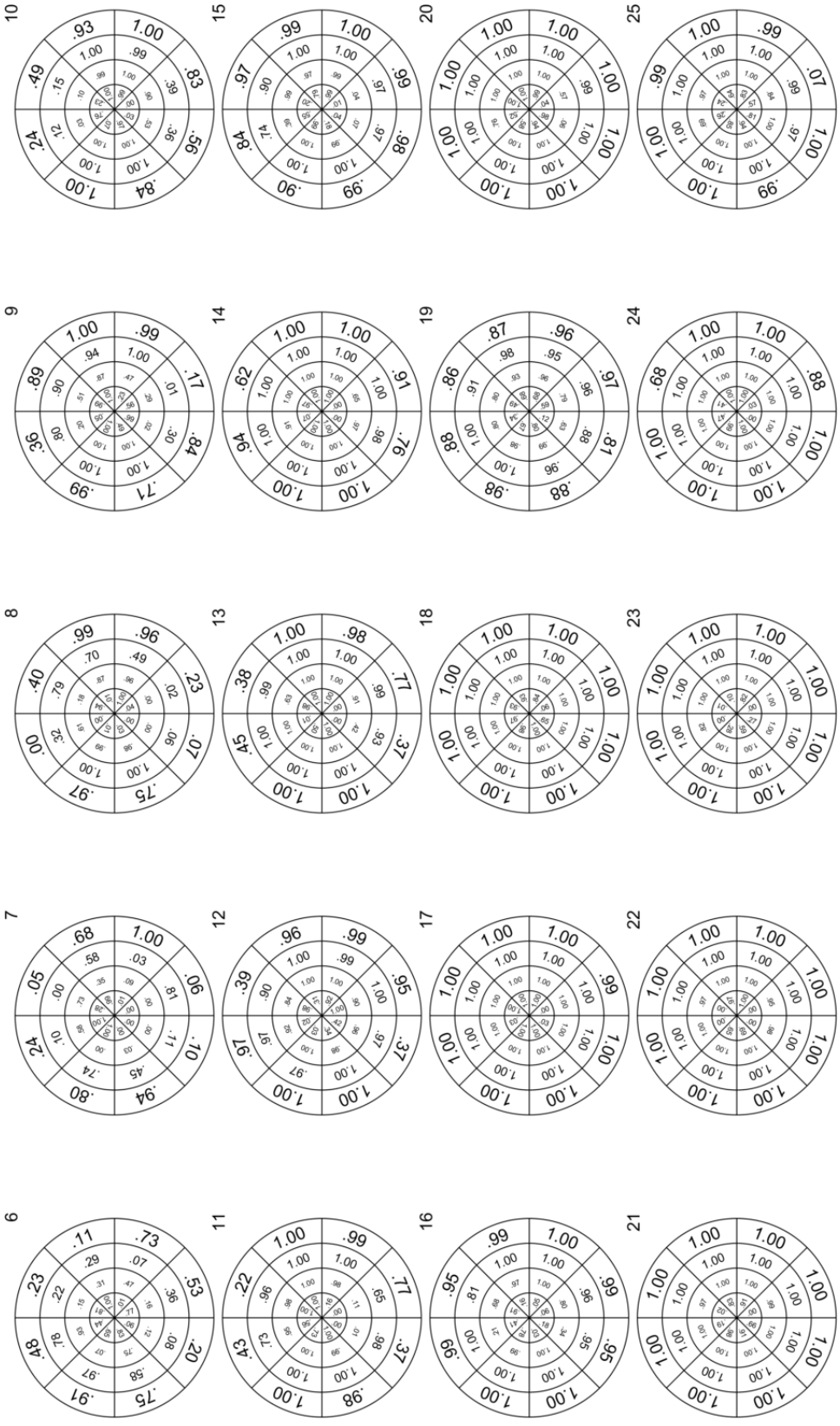

Fig. B.13 *Statistical Significance of Acceleration Behavior* P-values obtained from a left-tailed Monte Carlo test for the maps shown in Figure 2.6F

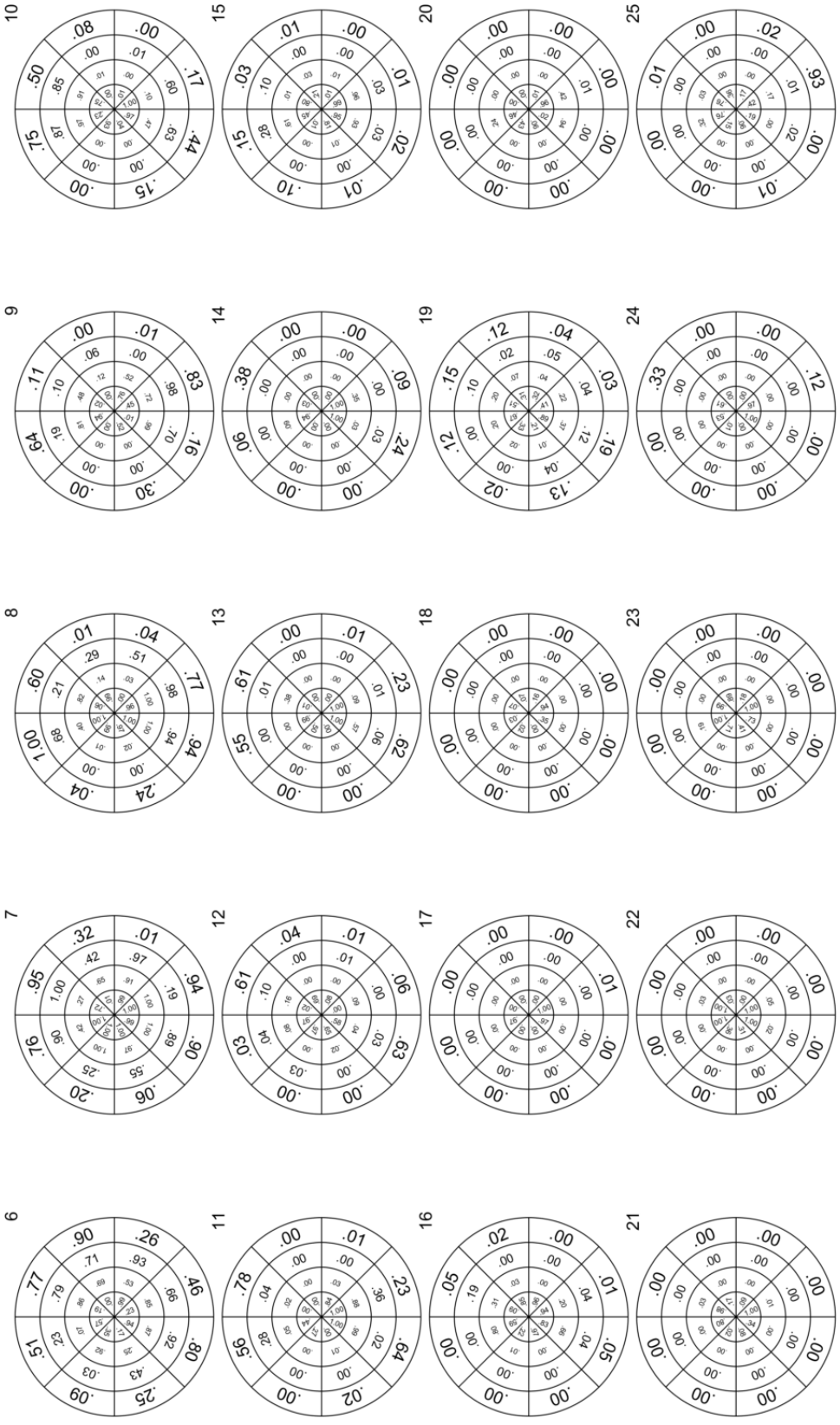

Fig. B.14 *Statistical Significance of Attraction* P-values obtained from a Monte Carlo test for the maps shown in Figure 2.7F